Universidad Europea

TRABAJO DE FIN DE GRADO

Grado en Odontología

CALCIFICACIONES CAROTÍDEAS EN ORTOPANTOMOGRÁFIAS

Madrid, curso 2020/2021

Número identificativo 11

RESUMEN

-Introducción: Mediante el uso habitual de las ortopantomografías en las clínicas dentales se puede ayudar al sistema de salud y a los pacientes enfermos en la prevención de accidentes cerebrovasculares causantes de una parte importante de los decesos en la población.

-Objetivos: Visualización, prevalencia, relaciones y factores de riesgo en relación con las calcificaciones carotideas en ortopantomografías.

-Material y método: El trabajo se ha realizó mediante el análisis de 61 estudios comprendidos entre los años 2002 y 2019, mediante el uso de plataformas digitales como PubMed, Sciielo, MedLine y ScienceDirect.

-Discusión: Tras la recopilación de la información mediante el análisis de los estudios referenciados comparamos todos los estudios para llegar a una cifra con el menor sesgo posible de placas de ateroma calcificadas en las radiografías panorámicas de las clínicas dentales.

-Conclusiones: Las radiografías panorámicas son un buen método para detectar placas de ateroma calcificadas en la arteria carótida.

ABSTRACT

-Introduction: With regular use of orthopantomography in the dental clinic, it helps patients and the health system to prevent cerebrovascular accidents that cause a significant amount of deaths in the population.

-Objetives: Visualization, prevalence, relationships and risks factors in relation to carotid calcifications in orthopantomography.

-Materials and methods: The study it has been made by analyzing 61 studies between the years 2002 and 2019, through the use of digital platforms such as PubMed, Sciielo, Medline and ScienceDirect.

-Discussion: After collecting the information by analyzing the referenced studies, we compared all the studies to arrive at a figure with the least possible bias of calcified atheroma plaques in panoramic radiographs of dental clinics.

-Conclusion: Panoramic radiographs are a good method for detect calcified atheroma plaques in the carotid artery.

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1 INTRODUCCIÓN

En algunas radiografías, los tejidos blandos presentan radiopacidades que pueden ser calcificaciones, osificaciones u objetos extraños (1). Se entiende por calcificación el deposito de sales de calcio que normalmente tiene lugar en los huesos y dientes; cuando ocurre en tejidos blandos se llama calcificación heterotrófica que a su vez se clasifican en 3 tipos: distróficas, idiopáticas y metastásicas (2). Las de tipo vascular son el objetivo de este estudio, y estas se clasifican dentro de las calcificaciones distróficas.

1.1 Mecanismos de calcificación vascular.

Las calcificaciones vasculares son un proceso activo que esta regulado y en el que intervienen diferentes mecanismos no excluyentes entre sí (3).

La calcificación ectópica es un mecanismo altamente regulado parecido a la ontogénesis. Existen diferentes similitudes entre el tejido vascular que se en cuenta calcificado y el hueso como por ejemplo la presencia de biopatitas, vesículas matriciales y de células que según condiciones en las que se encuentre, si son adecuadas pueden llegar a formar una matriz de mineralización (4,5)

1.2 Calcio y fosforo

Niveles elevados de Ca y F producen el crecimiento y formación de cristales de bioapatita (6). La bioapatita es el compuesto mineral principal en los huesos. Estudios sugieren que al exponer las CMLV al fósforo o calcio en altas concentraciones da lugar a la deposición de biopatita en la matriz extracelular y cuando se incuban los dos minerales al mismo tiempo se observa un efecto sinérgico de la calcificación (7). Aunque este proceso no es solo un mecanismo pasivo de precipitación de los iones, sino que implica diferentes procesos como un cambio fenotípico de las células del músculo liso vascular y de las up-regulation de los genes que se combina con la diferenciación osea. A partir de aquí se desarrollan una serie de mecanismos que actúan debido a la hiperfosfatemia y a la alta concentración de calcio y la asociación de estos factores lleva a la transformación de la célula para que se pueda llevar a cabo calcificación de la misma (8).

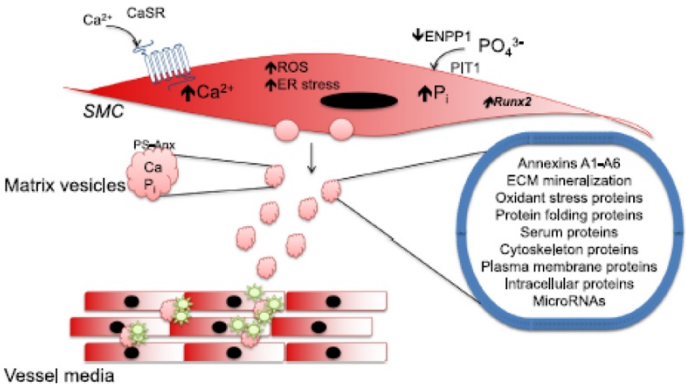

Figura 1. Efectos del fósforo y el calcio en la mineralización de las CMLV (9).

1.3 Muerte celular y apoptosis

Las llamadas vesículas matriz con contenido citoplasmático está en relación con la calcificación vascular. Dichas vesículas se sintetizan a raíz de las células donde se da origen a la mineralización o son producto de un procedimiento de apoptosis celular. El estudio realizado por Proudfoot et al. (10) muestra que la apoptosis regula in vitro la calcificación vascular. Conforme con el estudio de los autores mencionados, las vesículas de matriz pueden contener calcio en su interior y dan lugar a los cristales de biopatita.

1.4 Inhibidores de la calcificación

En condiciones normales las células vasculares cuentan con moléculas inhibidoras de la mineralización. La omisión de esta función en las células provoca la llamada 'perdida de la inhibición natural', y como resultado se llevará a cabo una calcificación espontanea y con ello la elevación de la mortalidad celular (11). A través de un análisis de mutaciones en animales se elaboró un dosier con las moléculas que inhiben los procesos de calcificación, entre las que incluimos:

1.4.1 Osteoprotegerina

Las células odontoblásticas sintetizan el factor diferenciador del osteoclasto ODF (RANKL), el cual se une al receptor de superficie del osteoclasto, dando lugar así a la agrupación y

posterior diferenciación de los precursores de los osteoclastos (OC) y procediendo a su activación e inhibición de la apoptosis (12). La osteoprotegerina, tiene origen en los odontoblastos y se une al RANKL comprometiendo la unión con el receptor osteoclástico (RANK). Así la OPG se considera un factor regulador óseo al actuar en competencia con RANK y finalmente proceder a la inhibición de la actividad osteoclástica (13).

1.4.2 Proteína Matrix Gla

La matrix gla es un inhibidor de la calcificación, el primero en ser identificado. Depende de la vitamina liposoluble K que se expresa en CMLV y en células del endotelio en vasos normales, pero su actividad se ve reducida en las arterias que se encuentran calcificadas.

1.4.3 Fetuína A

Es una Glicoproteina sérica y tisular que da lugar a la inhibición de la calcificación vascular ectópica. Inhibidor importante de la producción de hidroxiapatita, disminuyendo la síntesis de cristales que con calcio y fósforo in vitro (15)

1.4.4 Osteopontina

La osteopontina (OPN) pertenece a la familia SIBLING (proteínas N glucosiladas con ligando de unión de integrina) y a una glicoproteína N-Ligada. Estas glicoproteínas son sintetizadas por las células del hueso con actividad de conexión celular (16,17). Identificada por primera vez

en 1986 como una célula que se expresa en las células del sistema inmune y en las células del sistema vascular. La OPN juega un papel de mucha importancia en la regulación de la inflamación mediante la producción de citosinas en las células T activadas favoreciendo la concentración de monocitos/macrófagos

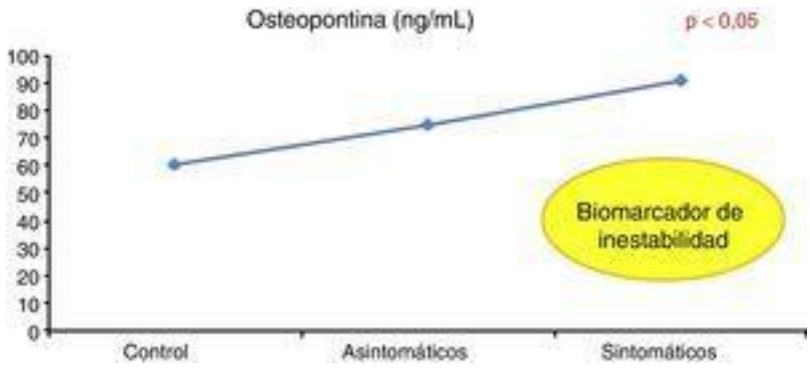

Figura 2 Aumento significativo de los niveles según el riesgo neurológico (18)

La expresión de la OPN en vasos sanguíneos y corazón en razonablemente baja, no obstante, se incrementa ante el daño y pronostica un funcionamiento cardiaco disminuido. Los niveles en plasma de OPN se correlacionan con el aumento y la extensión de la ateroesclerosis de las arterias coronarias (16). Por estas razones se sugiere que la OPN tiene relación con inflamación, calcificación y ateroesclerosis vasculares y que tiene importancia en el avance de la patología.

Así mismo, el estudio realizado por el Hospital Clínico de Valladolid y el Hospital 12 de Octubre de Madrid (19) tiene como finalidad la valoración de la OPN como marcador en la manifestación de la patología ateroesclerótica y su posterior acción perjudicial neurológica con la idea de usarla como marcador diagnóstico que ayude a la identificación de pacientes

asintomáticos. La hipótesis general de este estudio indica que existen diferencias en los niveles de inflamación en pacientes con síntomas y sin síntomas, aunque ambos padezcan ateroesclerosis carotídea.

1.4.5 Activadores de la calcificación

Hay estudios que indican que hay presencia de sustancias en el suero de pacientes con capacitación para estimular la calcificación (20):

·Fosfatasa alcalina: Se considera indispensable en el mecanismo de calcificación de las células vasculares. Su presencia ha sido detectada en calcificaciones vasculares y de válvulas cardiacas (21).

·Core binding factor alpha 1: esta sustancia es la reguladora principal de la diferenciación ósea. Los animales sometidos a estudios con carencia en Cbfa1 tienen dificultades para formar tejido cartilaginoso y mineralización del hueso (22).

·Bone morphogenic proteins (BMPs): Estas proteínas son llamadas de esta forma por sus características osteoinductivas. Los BMPs son de la familia del factor de crecimiento transformante beta (TGF-B). Desarrollan su acción uniéndose a receptores transmembrana que envían señales. La unión de una proteína BMP con su receptor da lugar a la activación de los receptores tipo I, lo que da lugar a la fosforilación y movimiento a otro espacio nuclear de factores de transcripción Smad cambiando de esta manera la tasa de transcripción de los genes diana (23). Promueven la síntesis ectópica de hueso (24)

Las calcificaciones vasculares se dividen principalmente en 2 grupos dependiendo de donde se deposite el fosfato de calcio en forma de cristales de biopatita (parecido al hueso) que se da tanto en vasos del sistema vascular como en válvulas del mismo sistema (25). En primer lugar las calcificaciones de la túnica intima, que está en relación con las placas de ateroma, y en segundo lugar las calcificaciones de la túnica media (conocida como esclerosis de Mönckeberg), relacionadas con el endurecimiento del sistema vascular debido a la mineralización de las fibras elásticas y la ateroesclerosis observada en pacientes de edad avanzada, enfermedad renal crónica y diabetes (26).

Las calcificaciones de la intima están ligadas a placas de ateroma debido a un crecimiento en los depósitos de lípidos y células inflamatorias en la túnica intima. Las calcificaciones de la túnica media tienen relación con el cambio de fenotipo en las células musculares lisas del tejido vascular hacia unas células mas semejantes a los osteoblastos, lo que supone una mayor rigidez en el tejido que forman (tejido vascular) (27).

1.5 Calcificaciones carotideas

Una placa de ateroma calcificada es una calcificación desarrollada en la túnica intima del tejido vascular que causa un mal funcionamiento en el sistema arterial a causa de la disminución de la luz de la arteria o vaso sanguíneo, produciendo así isquemia en los tejidos (28). Esta patología puede tener lugar en la arteria carótida común a la altura de su bifurcación entre las vértebras C4 y C5 en el lado izquierdo, en el lado derecho la calcificación tiene lugar entre la C3 y C4 (29). La evolución de placa de ateroma puede desencadenar una placa de ateroma inestable que se puede fracturar o ulcerar con la consiguiente posibilidad de obstruir la luz de

un vaso sanguíneo cerebral mas pequeño dando lugar a un accidente cerebrovascular (ACV) (30)(31).

El infarto cerebral o accidente cerebrovascular es un evento de isquemia mediante el cual la sangre no es capaz de llegar a una zona del cerebro a causa de un obstáculo, en este caso una placa de ateroma o calcificación vascular (32).

Las calcificaciones se pueden observar en radiografías panorámicas u ortopantomografias, técnica de imagen muy usada actualmente en la odontología (33). El deber del odontólogo en este caso es remitir a su medico de cabecera cualquier paciente con calcificaciones carotideas en las ortopantomografias para su evaluación (34).

1.6 Características Radiográficas

Para interpretar una radiografía panorámica hay que tener en cuentas varias cosas, como las densidades. Estas van desde radiopacidades muy heterogéneas con diferentes orientaciones hasta masas radiopacas en dirección al ángulo mandibular y el hueso hioides, por encima o por debajo de las vértebras cervicales 3 y 4 y superior al cartílago tiroides. (28) (35)

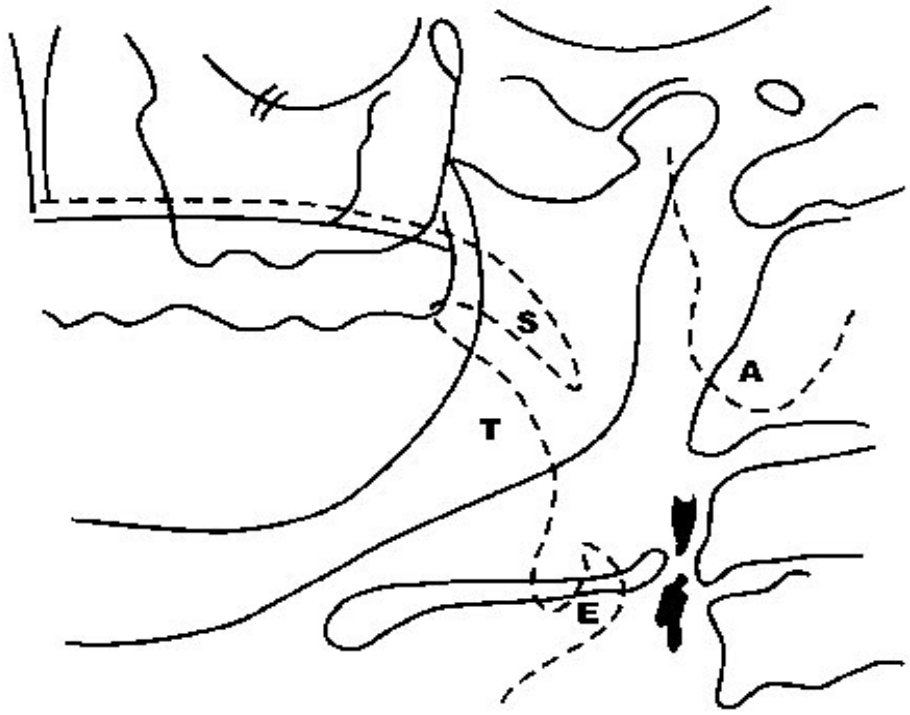

Figura 3. Ilustración de la placa de ateroma en la ortopantomografía. Fuente: Carter et al., (1997); p. 98215 (36)

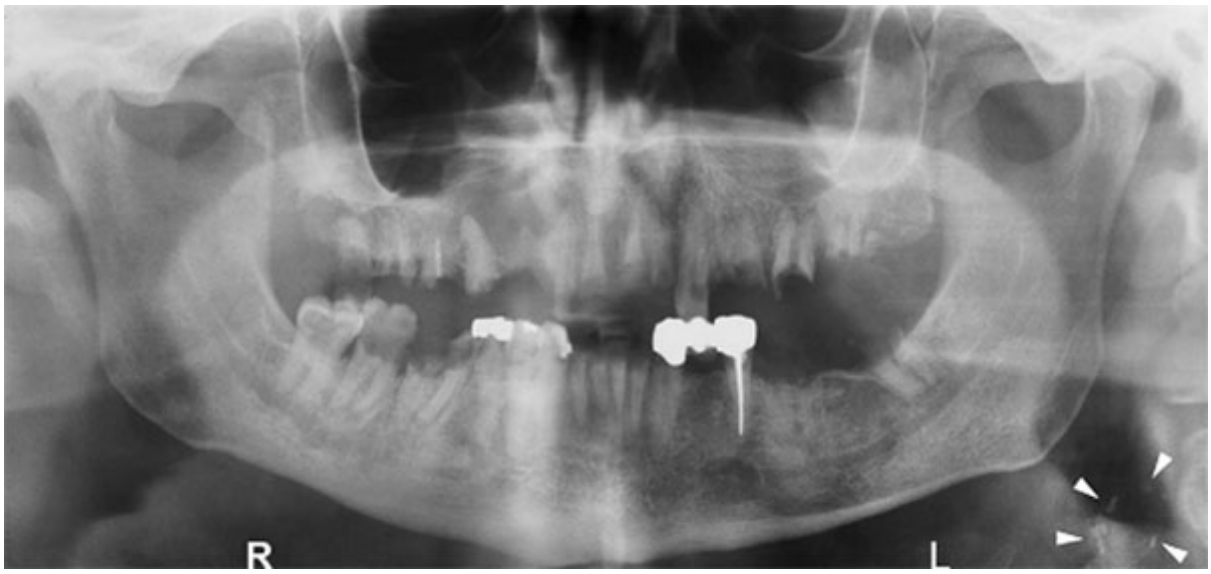

Figura 4. Las flechas indican la posición de las placas de ateroma

Fuente: Pornprasertsuk-Damrongsri et al, (2009), p. e5911. (35)

En una ortopantomografía o radiografía panorámica se muchos elementos anatómicos los cuales cualquier dentista debería saber. Se pueden apreciar inferior a la mandíbula a la altura vertical del segundo molar y a la altura en horizontal del mentón o se observan también en algunas ocasiones en la parte mas anterior de la radiografía la arteria carótida (no siempre se aprecia) (35).

Una vez detectada la figura extraña en una zona correspondiente a la carótida, derivaremos nuestro paciente a un especialista vascular para que pueda realizar pruebas mas concretas y por supuesto llevar a cabo un tratamiento que elimine el riesgo de sufrir un infarto cerebral y así evitar el riesgo de sufrir secuelas o fallecer (4).

1.7 Diagnóstico diferencial

En el diagnóstico diferencial de la calcificación carotideas se encuentran por un lado las entidades anatómicas y también las patológicas. Dentro de las anatómicas podemos destacar el cartílago triticio, el cuerno superior del cartílago tiroides, el asta mayor del hueso hioides, nódulos linfáticos calcificados y sialolitos pertenecientes a la glándula mandibular aunque cabe destacar también que la mayoría de ellos se encuentran en una parte mas superior que la placa calcificada (34).

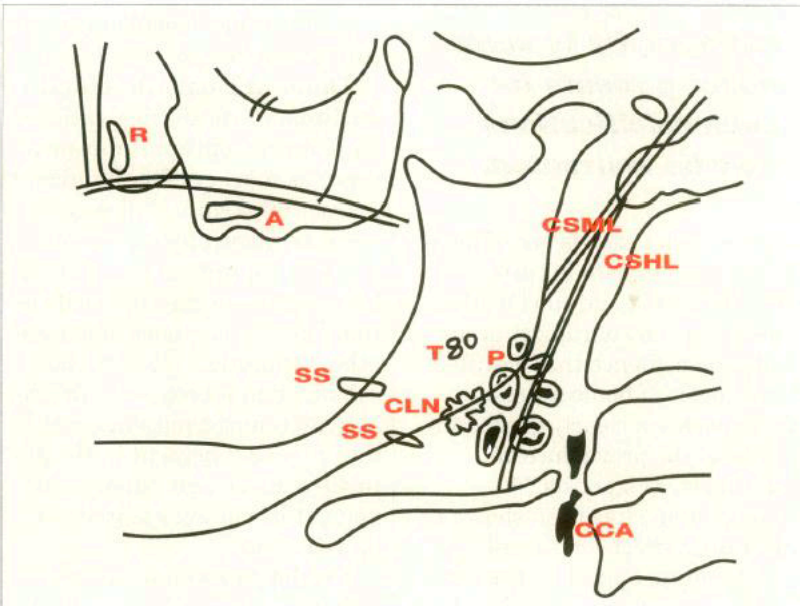

Figura 5. Ligamento estilomandibular calcificado, ligamento estilohioideo calcificado, sialolito submandibular(SS), flebolito(P), nódulo linfático calcificado(CLN) y arteria carótida calcificada(CCA) (36). Imagen de Carter et al.

Además, las entidades patológicas como los sialolitos que se hayan en la glándula submandibular, nódulos y flebolitos linfáticos calcificados. Los sialolitos suelen ser irregulares en cuanto a su forma, difusos y unilaterales, aparecen en radiografías panorámicas cerca del tercer molar inferior (35). Los flebolitos son ovalados con bordes alisados con una porción externa radiopaca y otra interna radiolúcida aunque también pueden ser totalmente radiopacos (37). Finalmente, los nódulos calcificados se localizan en la región submandibular y pueden afectar a un nódulo o varios de estos con apariencia de cadena, con borde definidos e irregulares (35) (38).

1.8 Entidades anatómicas:

-Cartílago tritíceo: se encuentran en la zona del esqueleto de la laringe. Esta estructura se encuentra en el ligamento tirohioideo lateral a la altura de la 3º y 4º vértebra. Calcificado, el cartílago tritíceo se puede apreciar de manera sencilla en una ortopantomografía y se puede diagnosticar como una calcificación de la arteria carótida u otras calcificaciones de tejidos blandos (39).

Hueso hioides: su imagen en la radiografía panorámica es bilateral, con un incremento del formato vertical, nada bien definida, en el ángulo posterior mandibular, superior al cuerno mayor del hioides (40).

La epíglotis: con forma de media luna u hoja vertical que aparece detrás de la lengua y hacia arriba, encima del cuerno mayor del hioides y posterior al ángulo de la mandíbula, controlando los flujos de aire y comida que pasan por la faringe(35).

1.9 Factores de riesgo

Los estudios y datos experimentales muestran que es un mecanismo que se desencadena por varios factores de riesgo entre los que se aprecian la hipertensión arterial (HA), el envejecimiento celular, la dislipidemia, diabetes milletus, el estrés oxidativo, fumar tabaco, etc (42). Debido a estos factores denominados de riesgo, se dará lugar a la inflamación y posterior activación del factor nuclear kB. Este, es clave en los mecanismos fisiológicos de supervivencia celular, proliferación y activación (43).

1.10 Ortopantomografía

Si bien es cierto que la ortopantomografia o radiografía panorámica no es la opción mas adecuada para detectar calcificaciones carotideas, también es cierto que muchos estudios han avalado su utilidad no solo para uso odontológico sino para hallazgos accidentales como las calcificaciones en la región cervical (36) (41).

El papel del dentista en la detección de placas calcificadas a través de ortopantomografias, historial medico, factores de riesgo, etc, cada vez es de mas importancia debido a la gran ventaja que supone el diagnostico precoz de esta enfermedad (44).

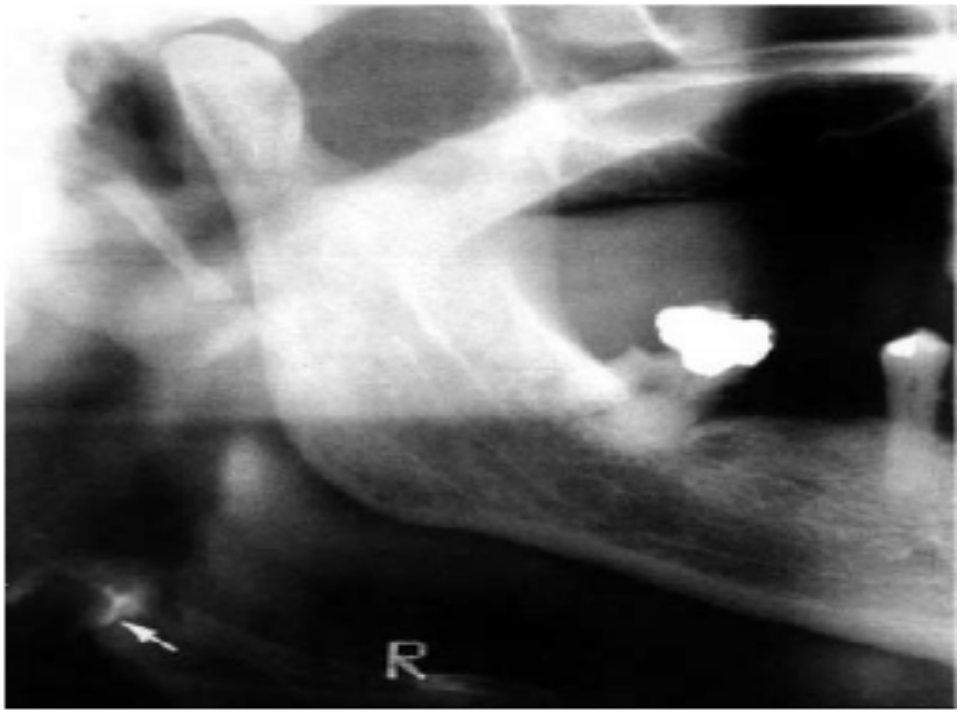

Figura 6. Ortopantomografía recortada digitalmente para apreciar el proceso ateroesclerótico carotideo. La flecha indica la calcificación. (Dr Arthur H. Friedlander)

En los años 80 (45) se investigo una circunstancia que podría acelerar el diagnóstico precoz de las placas de ateroma en la arteria carótida. Esta investigación se basa en que el 85% de los Accidentes Cerebrovasculares tiene origen isquémico, de los cuales se piensa que el 65% de ellos esta causado debido a la formación de trombos y émbolos en la región de la bifurcación de la arteria carótida. Este autor (45) declara la importancia de las radiografías panorámicas para la detección de estas calcificaciones a nivel de la bifurcación carotidea que pasaran a los vasos sanguíneos cerebrales sin esa detección precoz.

Otro punto a tener en cuenta en la posición en contra de algunos autores a la hora de la realización de placas panorámicas para la detección de ateroesclerosis debido a la poca exactitud de la imagen, la cual no permite llegar a un diagnóstico certero, pero si permite que sea una opción mas en nuestro diagnostico diferencial, el cual terminara de ejecutar el medico, ya nombrado anteriormente (45), encargado de tratar este tipo de patologías (46).

2 OBJETIVOS

1- Comprobar si las calcificaciones carotídeas pueden ser visualizadas por el odontólogo en las evaluaciones dentales rutinarias.

2- Establecer cuál es la prevalencia de las calcificaciones carotídeas en los diferentes estudios analizados.

3- Comprobar si existe una predilección por la presentación unilateral o bilateral.

4- Establecer si existe relación entre las calcificaciones carotídeas y la edad, sexo y factores de riesgos cardiovasculares.

3 METODOLOGÍA

Para este estudio, se ha realizado una búsqueda bibliográfica en Pub-Med/ MEDLINE empleando las palabras clave: « Carotid Calcificated in panoramics », « Anatomy of Carotid artery », « Carotid artery atheromas », « Ortopantomography », « Ateroma », « Carotid stenosis ».También se realizado una búsqueda en google con las siguientes palabras clave: « Carotid Calcificated in panoramics », « Diagnostic of Carotid Calcificated in panoramics ».

En este trabajo se han analizado 26 estudios de los veinte últimos años sobre la prevalencia de la presencia de estas calcificaciones dependiendo de la edad, el sexo y de los riesgos de enfermedades cardiovasculares (hipertensión, obesidad, hipercolesterolemia, diabetes, fumador y antecedentes de infartos) y 35 estudios relacionados con la anatomía, radiografías panorámicas, bioquímica en relación a las calcificaciones y placas de ateroma, características radiográficas También se ha realizado una búsqueda de artículos científicos en los que se concluyeran datos acerca de la localización de las calcificaciones (Unilateral o bilateral, derecho o izquierdo). Otros artículos han ayudado al presente trabajo de fin de Grado para definir y describir la patología así como para las ilustraciones empleadas en el mismo.

-Como criterios de inclusión:

-Estudios con tamaño muestral significativo.

-Estudios con una buena descripción clínica y anatómica de los procesos.

-Artículos con buenas imágenes.

-Como criterios de exclusión:

-Estudios sin significación estadística.

-Estudios anteriores a 2001.

4 RESULTADOS

Tras aplicar los criterios mencionados en el apartado de metodología, la búsqueda resultó en un total de 23 artículos resumidos en una tabla en la que exponen los siguientes datos: Autores, artículo, nº de radiografías estudiadas, incidencia de calcificaciones, sujetos, edad y sexo.

Algunos artículos además de tener en cuenta las calcificaciones carotideas (grupo experimental) también tienen un grupo control para poder determinar la relación de incidencia de calcificaciones de una determinada enfermedad como la diabetes o la relación existente entre las calcificaciones carotídeas y los accidentes cerebro-vasculares.

Los artículos comprenden desde los 4 años hasta los 97 años y en la mayoría de los casos la población es de ambos sexos.

Autores	Artículo	Nº de radiografías estudiadas	Incidencia de calcificaciones	Sujetos	Edad	Sexo
V. E. Rushton, et al	La ortopantomografia como método para la detección de las placas de ateroma calcificadas	1818	3,1%	1818	Mayores de 18 años	Hombres y mujeres
N. Cohen, et al	*Carotid calcification on panoramic radiographs: An important marker for vascular risk*	1879	3,8%	1879	Mayores de 55	Hombres
Cristina Barona-Dorado, et al	*Relation between diagnosis of atheromatous plaque from orthopantomographs and cardiovascular risk factors*	1602	15,4%	1541	Entre 18 y 97 años	897 hombres y 705 mujeres
Dov. Almog, et al	*Evaluation of a training program for detection of carotid artery calcifications on panoramic radiographs*	778	3,47%	778	Media de 67 años	298 hombres y 480 mujeres
Takeshi Ohba, et al	*Evaluation of calcified carotid artery atheromas detected by panoramic radiograph among 80-year-olds*	659	5%	659	> de 80 años	262 hombres y 397 mujeres
T. Tamura, et al	*Clinicostatistical study of carotid calcification on panoramic radiographs*	2568	4,1%	2568	Entre 50 y 70 años	Hombres y mujeres

Stefan Bayer, et al	*Prevalence of findings compatible with carotid artery calcifications on dental panoramic radiographs*	2557	4,8%	2557	> de30 años	1507 mujeres y 1048 hombres
Alven Arreaza, et al	Ateroma calcificado en la carótida y radiografía panorámica: reporte de caso	1	100%	1	49 años	Mujer
Guerreiro N, et al	*Prevalence of calcified carotid artery atheromas in panoramic radiographs of HIV-positive patients*	300	8,2%	300	> de 18 años	Hombres y mujeres
AH. Friedlander, et al	*Panoramic radiographic identification of carotid arterial plaques*	1000	2%	1000	Entre 50 y 75 años	Hombres y mujeres
S. Pornprasertsuk-Damrongsri, et al	*Carotid artery calcification detected on panoramic radiographs in a group of Thai population*	1370	2,5%	1370	Entre 50 y 87 años	Hombres y mujeres
N. Khambete, et al	*Reliability of digital panoramic radiographs in detecting calcified carotid artery atheromatous plaques: A clinical study*	100	37%	50	Entre 50 y 84 años	Hombres y mujeres

AH. Friedlander, et al	*The prevalence of calcified carotid artery atheromas on the panoramic radiographs of patients with type 2 diabetes mellitus*	49	Diabeticos 20,4% Grupo control 4%	49	Entre 55 y 81 años	Hombres
Y. Sisman, et al	*The Prevalence of Carotid Artery Calcification on the Panoramic Radiographs in Cappadocia Region Population*	750	5,06%	1282	> de 40 años	482 Hombres y 268 mujeres
F. Ezoddini-Ardakani, et al	*Evaluation of positive predictive value for digital panoramic radiography in comparison to ultrasound in diagnosis of calcified carotid atheroma*	1682	2,43%	1682	Entre 22 y 62 años	16 hombres y 27 mujeres
CM. Romano-Sousa, et al	*Diagnostic agreement between panoramic radiographs and color doppler images of carotid atheroma*	32	3,1%	16	Sin especificar	Sin especificar

J. Gonçalves, et al	*Prevalence of pathologic findings in panoramic radiographs: Calcified carotid artery atheroma*	8338	6,9%	8338	Entre 4 y 94 años	5049 mujeres y 3289 hombres
M. Imanimoghaddam	*Dopple sonography confirmation in patients showing calcified carotid artery atheroma in panoramic radiography*	960	1,97	960	> de 40 años	524 mujeres y 436 hombres
B. Baryam, et al	*Digital panoramic radiography: a reliable method to diagnose arotid artery atheromas?*	4106	-2,1% -75,1% mujeres y 24,9% hombres	4106	> de 40 años	2428 mujeres y 1678 hombres
S-O. Aranzazu, et al	Detección de placas de ateroma mediante radiografías dentales	459	2,83% de los pacientes (53,84% hombres y 46,15% mujeres)	1300	> de 40 años	194 hombres y 265 mujeres
Elias Johansson MD, et al	*Carotid calcifications on panoramic radiographs: a 5-years follow up study.*	1183	9,5%	1182	Edad entre 18 y 74 años	Hombres y mujeres
I. Nasseh, et al	*Carotid artery calcification: A digital panoramic-Based stufdio*	500	6,8%	500	281 mujeres y 219 hombres	Edad entre 18 y 88 años
R. Tiller, et al	*Association between carotid area calcifications and periodontal risk: a cross sectional study of panoramic radiographic findings*	1521	9.0%	824	349 hombres y 475 mujeres	Edad entre 65 y 32 años

5 DISCUSIÓN

Las arterias carótidas en su zona común se diferencian en dirección, trayecto, longitud y relaciones y esto es porque no tienen igual origen. Por una parte, la carótida común derecha se origina en el tronco braquiocefálico y se encuentra en la parte anterior del cuello y por otra parte la carótida común izquierda tiene su nacimiento en el propio cayado aórtico (motivo por el que la izquierda es mas larga que la carótida derecha), de ahí que su origen se sitúe mas alejado de el encéfalo. Desde ahí surcan un trayecto ascendente hasta que se bifurcan. Esta bifurcación tiene lugar a 1 del cartílago tiroides, en concreto a 1 centímetro de su borde superior (47).

Las placas calcificadas dan lugar a una disminución de la luz del vaso y debido a esto la cantidad de sangre que fluye hasta su zona de irrigación es menor de la que debería, lo cual pone en riesgo la salud y vida del paciente (30) (31), dando lugar a una enfermedad ateroesclerótica que causa una parte muy importante de las muertes mundiales (48). He aquí la importancia del diagnostico precoz de este tipo de enfermedades.

Para el correcto diagnóstico de las placas calcificadas carotideas Carter et al (36) nos facilita datos muy importantes para poder diferenciar la placa calcificada de otras patologías en la misma región. Una placa calcificada se visualiza como una imagen radiopaca irregulares, verticolineales, heterogéneas ubicadas en la zona inferior del ángulo de la mandíbula y a un lado de las vertebras cervicales 3 y 4, laterales a la ubicación de el cartílago tritíceo y el cuerno superior del cartílago tiroides (49).

En el año 1981 Arthur Friedlander et al (45) enseño al mundo la presencia de estas calcificaciones en los tejidos blandos del cuerpo humano mediante ortopantomografías (50). Desde ese año se han realizado numerosas investigaciones y estudios que reportan entre un 2-5% de prevalencia de calcificaciones en la OPG de pacientes sin enfermedades sistémicas (29) (35), sin embargo, un estudio realizado por Khambete et al (51) asegura que la incidencia era del 34% lo cual algunos autores lo atribuyeron a usar radiografías digitales, concluendo asi que las OPG tenían una sensibilidad del 76% y una especificidad del 99% para localizar las placas de ateroma calcificadas. Por otra parte, el análisis de las OPG en la población con algún factor de riesgo fue mas elevado, llegando a un intervalo entre 20% y 38% de frecuencia de calcificaciones (52). Entre los factores de riesgo, los autores clasificaron como factores no modificables la edad, y a los modificables a las enfermedades sistémicas y hábitos nocivos como por ejemplo la HA, hiperlipidemia, diabetes, fumadores, etc.

En 2003 Ohba et al (29) reporta una frecuencia del 5% en una muestra poblacional de casi setecientas personas sin síntomas, resultados muy parecidos a la investigación realizada por Tamura et al (48). El estudio de Sismas (53) et al y Bayer et al (34) dan un 4.13 y un 5.06 respectivamente.

La gran parte de estudios sobre esta materia están de acuerdo en que la frecuencia es de entre 2 y 5 por cierto en pacientes sin factores de riesgo.

Pero, ¿cómo de fiable es la información de una OPG en el hallazgo de las calcificaciones?.

Según el estudio de investigación realizado por Ezoddini-Ardakani et al (54) concluyen que se puede considerar de utilidad en el diagnoótico precoz de calcificaciones carotideas y se basan por ejemplo en que las visitas al dentista se suelen dar con mas frecuencia y la realización de OPG por consiguiente es mas común. Por otra parte, Romano-Sousa et al (55) determina un nivel alto de coincidencia en la detección de placas calcificadas carotideas entre una OPG y la técnica de imágenes doppler.

En cuanto a la presencia de factor de riesgo inmodificable por la condición sexual el sexo femenino tenia mas calcificaciones que el masculino (29) (48) (56).

El estudio que realizó obha et al (29) el sexo mas afectado era el femenino, Tamura et al(48) dio a además el ratio hombre mujer que fue de 1:3,07.

En cuanto a la relación entre la edad y el sexo en los pacientes con calcificaciones carotideas se observó que en el estudio realizado por Golçalves et al (56) había una mayor frecuencia en el intervalo de edad entre 40 y 70 años interpretando así que se dan valores suficientes como para hacer una asociación entre las ACAV (ateroma calcificado de arteria carótida) y la edad, sin embargo para Ezoddini-Ardakani et al (54) fue mas amplio el rango, de los 22-62.

Los estudios realizados indican que la capacidad de sufrir ACAV aumenta con la edad (46) ya que con la edad se hacen cada vez mas notables los factores que contribuyen a la formación de placas calcificadas en la arteria carótida, sin embargo, con cambios de la población general en hábitos de estilo de vida se puede reducir mucho el riesgo de aparición de una ACAC.

Según la gran mayoría de los estudios realizados la placa calcificada en la arteria carótida se presento de manera unilateral con una relación 2:1, siendo 1 las que se presentan de forma

unilateral frente a las bilaterales (50) (53). Sin embargo el estudio realizado por Imanmoghaddam et al (57) asegura haber obtenido cifras muy diferentes a los de los anteriores estudios: 66% bilaterales, el doble que las unilaterales contradiciendo así la tendencia de los demás estudios. Por otro lado, Tamura et al (48) no indica ninguna diferencia entre presencia de calcificaciones bilaterales o unilaterales.

En cuanto a la ubicación en lo que al lado del cuerpo se refiere el lado izquierdo es el que coincide con la mayoría de los estudios (48) (50) (56) (57) (58). No obstante Tamura et al (48) determina que la prevalencia de calcificaciones tanto en hombres como en mujeres en el lado izquierdo fue levemente superior a la del lado derecho, contradiciendo la conclusión al respecto de Sisman et al (53) y ohba et al (29) los cuales obtuvieron un mayor porcentaje en el derecho.

En el año 2000 (28) se realizo un estudio realmente interesante en el que se trato de averiguar si la incidencia de las lesiones calcificadas de la arteria carótida observadas en una OPG podría variar si el observador fuera un cirujano o maxilofacial o un dentista estomatólogo general. Estudios anteriores situaban la prevalencia entre el 2-5% (34) (48) (50) (54) (57) (58) en las calcificaciones carotideas observadas en OPG en pacientes dentales ambulatorios. A partir de aquí se elaboro un estudio para determinar la variabilidad en los diferentes examinadores para la detección de ACAC en OPG. Los dentistas estomatólogos fueron entrenados para observar este tipo de patologías en OPG mediante el estudio de la anatomía regional y los diferentes diagnostivos diferenciales a través de una academia, la Academia Americana de Radiología Oral y Maxilofacial (AAROM). Los resultados de este estudio son bastante contundentes. El primer observador de la OPG, un graduada en odontología residente de

segundo año identifico 99 personas de una muestra de 778 con supuestas ACAV, lo que sería una prevalencia del 12%. El segundo examinador es un docente de prostodoncia el cual también recibió entrenamiento por parte de la academia (AAROM) e identifico 78 casos en los 99 casos que el primer examinador examinó, bajando así la prevalencia en 2 puntos hasta un 10%. Finalmente el examinador final que fue el que impartió el curso de la academia Americana, profesor de ciencias diagnoticas orales, reviso las 78 radiografías hasta llegar a los 27 individuos con calcificaciones en la arteria carótida lo que hace tener una prevalencia del 3,5 % (Friedlander).

Las calcificaciones tanto cartílago tritíceo como de otros cartílagos laríngeos supusieron el 82% de los errores en cuanto a malas interpretaciones (28). Según Almong et al (28) hace falta una cualificación mediante el aprendizaje para poder detectar calcificaciones carotideas de manera certera, una buena ayuda podría ser medico radiólogos especializados en la región maxilofacial.

El cartílago tritíceo, objeto de error en la identificación de las placas calcificadas aparece medial a la placa calcificada carotídea (36). Aun así, hará falta el uso de otros diagnósticos de imagen como Doppler o tomografía computerizada para llegar a un diagnostico certero.

En cuanto al tratamiento el mejor para ACAC es du detección precoz. Los principales métodos de diagnostico son el doppler de ultrasonidos y la angiografía (35).

En 1991 se publico el North American Symptomatic Endarterectomy Collaborators (NASCET) en el cual la mitad de 659 personas con una obstrucción mayor de 70% de la luz del vaso recibieron tratamiento medico y la otra mitad recibieron una cirugía endarterectomía

carotidea (59). Este estudio fue suspendido debido a que un análisis a lo largo del curso determinó la clara mejoría de los pacientes con la cirugía.

En los pacientes en los que la estenosis no llega al 70% se observa que los que tienen una estenosis entre el 50 y 69% se aprecian evolución claramente favorable.

Desde el año 1953 De Bakey llevó a cabo la primera endartectomía carotidea y a día de hoy se han realizado un millón de estas intervenciones en el mundo (60). En EE.UU. el numero de cirugías carotideas aumento a 107 mil en 1985 (61). Luego disminuyó las cirugías hasta 70 mil en el año 1990.

6 CONSLUSIONES

-Las calcificaciones carotídeas pueden ser observadas en exámenes odontológicos rutinarios en los que se utilicen ortopantomografías

-Haciendo la media de todos los estudios analizados en los que no se examinan relaciones con otras patologías, es decir, entre la población estándar, la prevalencia de calcificaciones carotideas observadas en radiografías panorámicas es del 4,34%.

-Aunque no todos los estudios diferencian entre calcificaciones unilaterales o bilaterales en la mayoría de los estudios que las incluyen concluyen que tienen mas probabilidad de ser unilaterales frente a bilaterales.

-Según los estudios realizados y posteriormente analizados podemos concluir que las mujeres son mas propensas a desarrollar calcificaciones carotideas frente a los hombres. También en la edad comprendida entre los 55 y 75 años observamos un aumento considerable de los casos de ateroesclerosis carotidea. Finalmente, el RR (riesgo relativo) de personas que cursan con placas de ateroma calcificadas y su relación con factores de riesgo cardiovasculares es positiva, siendo RR>1.5.

7 BIBLIOGRAFÍA

1. Noffke CEE, Raubenheimer EJ, Chabikuli NJ. Radiopacities in soft tissue on dental radiographs: Diagnostic considerations. S Afr Dent J. 2015;70(2):53-7.

2. Vengalath J, Puttabuddi JH, Rajkumar B, Shivakumar GC. Prevalence of soft tissue calcifications on digital panoramic radiographs: A retrospective study. J Indian Acad Oral Med Radiol. 2014;26:385-9.

3. Speer MY, Giachelli CM. Regulation of cardiovascular calcification. Cardiovascular Pathology 2004;13:63–70.

4. Speer M, Giachelli C. Regulation of cardiovascular calcification. Cardiovasc Pathol. 2004; 13 (2):63-70.

5. London G. Mechanisms of arterial calcifications and consequences for cardiovascular function. Kidney Int Suppl. 2013; 3(5):442-5.

6. Block GA, Port FK. Re–evaluation of risks associated with hyperphosphatemia and hyperparathyroidism in dialysis patients: Recommendations for a change in management. Am J Kidney Dis 2000;35:1226–37.

7. Reynolds JL, Joannides AJ, Skepper JN, et al. Human vascular smooth muscle cells undergo vesicle–mediated calcification in response to changes in extracellular calcium and phosphate concentrations: A potential mechanism for accelerated vascular calcification in ESRD. J Am Soc Nephrol 2004;15:2857–67.

8. Steitz SA, Speer MY, Curinga G, et al. Smooth muscle cell phenotypic transition associated with calcification: upregulation of Cbfa1 and down regulation of smooth muscle lineage markers. Circ Res 2001;89:1147–54.

9. Karwowski, W., Naumnik, B., Szczepański, M., & Myśliwiec, M. (2012). The mechanism of vascular calcification – a systematic review. Medical Science Monitor, 18(1), RA1-RA11.

10. Proudfoot D, Skepper JN, Hegyi L, et al. Apoptosis regulates human vascular calcification in vitro – Evidence for initiation of vascular calcification by apoptotic bodies. Circ Res 2000;87:1055–62.

11.

12. Lacey DL, Timms E, Tan H-L, Kelley MJ, Dunstan CR, Burgess T, et al..

Osteoprotegerin (OPG) ligand is a cytokine that regulates osteoclast differentiation and activation..

Cell, 93 (1998), pp. 165-76

13. Tyson KL, Reynolds JL, McNair R, et al. Osteo/chondrocytic transcription factors and their target genes exhibit distinct patterns of expression in human arterial calcification. Arterioscler Thromb Vasc Biol 2003;23:489–94.

14. Heiss A, DuChesne A, Denecke B, et al. Structural basis of calcification inhibition by alpha 2–HS glycoprotein/fetuin–A. Formation of colloidal calciprotein particles. J Biol Chem 2003;278:13333–41.

15. J.L. Coloma, C. Ros, C. Castelo-Blanco.

Osteoporosis. Fisiología y patología-1.

REVCOG., 14 (2009), pp. 178-187

16. V. Lamour, M-J. Nokin, A. Henry, V. Castronovo, A. Bellahcène.

SIBLING proteins: Molecular tools for tumor progression and angiogenesis.

Médecine Sci MS., 29 (2013), pp. 1018-1025

17. González Fajardo J, Zamora González N, Aguirre Gervás B, Marcos Sánchez H, Vidriales Vicente I, Calvo Nieves M, Vaquero Puerta C. La Osteopontina como biomarcador de riesgo neurológico en la enfermedad carotídea. [Internet]. 2016 [citado 10 enero 2021];. Disponible

en: https://www.elsevier.es/es-revista-angiologia-294-articulo-la-osteopontina-como-biomarcador-riesgo-S0003317016300608

18. H-J. Cho, H-J. Cho, H-S. Kim. Osteopontin: A multifunctional protein at the crossroads of inflammation, atherosclerosis, and vascular calcification. Curr Atheroscler Rep., 11 (2009), pp. 206-213

19. Giachelli CM. Vascular calcification mechanisms. J Am Soc Nephrol 2004;15:2959–64.

20. Moe SM, Duan D, Doehle BP, et al. Uremia induces the osteoblast differentiation factor Cbfa1 in human blood vessels. Kidney Int 2003;63:1003–11.

21. Shioi A, Katagi M, Okuno Y, et al. Induction of bone–type alkaline phosphatase in human vascular smooth muscle cells: roles of tumor necrosis factor–alpha and oncostatin M derived from macrophages. Circ Res 2002;91:9–16.

22. Ducy P, Zhang R, Geoffroy V, et al. Osf2/Cbfa1: a transcriptional activator of osteoblast differentiation. Cell 1997;89:747–54.

23. Chen D, Zhao M, Mundy GR. Bone morphogenetic proteins. Growth Factors 2004;22:233–41

24. Wang EA, Rosen V, D'Alessandro JS, et al. Recombinant human bone morphogenetic protein induces bone formation. Proc Natl Acad Sci USA 1990;87:2220–4.

25. Edmonds ME, Morrison N, Laws JW, Watkins PJ. Medial arterial calcification and diabetic neuropathy. Br Med J (Clin Res Ed) 1982;284:928–30.

26. Valdivielso, Jose & José, Correspondencia & Valdivielso, Manuel. (2011). Calcificación vascular: tipos y mecanismos.

27. P. Lanzer, M. Boehm, V. Sorribas, M. Thiriet, J. Janzen, T. Zeller, *et al*. Medial vascular calcification revisited: Review and perspectives. Eur Heart J., 35 (2014), pp. 1515-1525

28. Almog D, Tsimidis K, Moss M, Gottlieb R, Carter L. Evaluation of a training program for detection of carotid artery calcifications on panoramic radiographs. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2000; 90:111-7.

29. Ohba T, Takata Y, Ansai T, Morimoto Y, Tanaka T, Kito S, et al. Evaluation of calcified carotid artery atheromas detected by panoramic radiograph among 80-year-olds. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2003; 96(5):647-50.

30. Madden R, Hodges J, Salmen C, Rindal D, Tunio J, Michalowicz B, Ahmad M. Utility of panoramic radiographs in detecting cervical calcified carotid atheroma. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2007; 103(4):543-8.

31. Ogata J, Masuda J, Yutani C, Yamaguchi T. Rupture of atheromatous plaque as a cause of thrombotic occlusion of stenotic internal carotid artery. Stroke. 1990; 21(12):1740–5.

32. Accidente cerebrovascular: Esperanza en la investigación [Internet]. National Institute of Neurogical Disorders and Strokes. 2000 [citado 2 febrero 2021]. Disponible en: https://espanol.ninds.nih.gov/trastornos/accidente_cerebrovascular.htm

33. Rushton VE, Horner K, Worthington HV. Screening panoramic radiology of adults in general dental practice: radiological findings. Br Dent J 2001;190:495-501.

34. Bayer S, Helfgen E, Bös C, Kraus D, Enkling N, Mue S. Prevalence of findings compatible with carotid artery calcifications on dental panoramic radiographs. Clin Oral Invest .2011; 15:563–9.

35. Friedlander A, Gratt B. Panoramic dental radiography as an aid in detecting patients at risk for stroke. J Oral Maxillofac Surg. 1994; 52(12):1257-62.

36. Carter LC, Haller AD, Nadarajah V, Calamel AD, Aguirre A. Use of panoramic radiography among an ambulatory dental population to detect patients at risk of stroke. J Am Dent Assoc. 1997;128:977-984.

37. Monsour P, Romaniuk K, Hutchings R. Soft tissue calcifications in the differential diagnosis of opacities superimposed over the mandible by dental panoramic radiography. Aust Dent J. 1991; 36(2):94-101.

38. Langlais R, Langland O, Nortjé C. Soft tissue radiopacities. En: Beaulieu M/Zinner S/ Dirienzi S. Diagnostic imaging of the jaws. 1st ed. Baltimore: Williams &Wilkins; 1995. 617-

39. Kamikawa Rss. Pesquisa de calcificaçoes em tecidos moles na regiao cervical por meio das técnicas radiográficas panorámica d telerradiografía. [tese]. Sao Paulo: universidade de Sao Paulo; 2009.

40. Arreaza A, Lara M. Ateroma calcificado en carótida y radiografía panorámica: reporte de caso [Internet]. Actaodontologica.com. 2020. Available from: https://www.actaodontologica.com/ediciones/2011/3/art-15/

41. Guerreiro N, Nelo E, Mesquita F, Warmling L,Ortega K. Prevalence of calcified carotid artery in panoramic radiographs of HIV- positive patients undergoing antirretroviral treatment: a retrospective study. Oral bSurg Oral Pathol Oral Radiol 2014; 117: 67-74.

42. Stocker R, Keaney J. Role of oxidative modifications in atherosclerosis. Physiol Rev. 2004; 84(4):1381-478. 43.

43. Shao J, Sadhu J, Towler D. Inflammation and the osteogenic regulation of vascular calcification. A review and perspective. Hypertension. 2010; 55(3): 579–92.

44. Little JW, Falace DA, Miller CS, Rhodus NL, eds. Dental management of the medically compromised patient. St. Louis: Mosby; 2002. p. 417-38.

45. Friedlander AH, Baker JD. Panoramic Radiography: an aid in detecting patients at risk of cerebrovascular accident. J Am Dent Assoc 1994;125:1598-603.

46. Friedlander A, Altman L. Carotid artery atheromas in postmenopausal women: Their prevalence on panoramic radiographs and their relationship to atherogenic risk factors. J Am Dent Assoc. 2001; 132(8): 1130-6.

47. Rouviere H. Vasos de la cabeza y del cuello. En: Gotzens V/ Finel G. Anatomía Humana descrptiva, topográfica y funcional. 11° Edic. Barcelona: Masson; 2005. 214 - 271.

48. Tamura T, Inui M, Nakase M, Nakamura S, Okumura K, Tagawa T. Clinicostatistical study of carotid calcification on panoramic radiographs. Oral Dis. 2005; 11(5):314-7.

49. Carter L. Discrimination between calcified triticeous cartilage and calcified carotid atheroma on panoramic radiography. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2000; 90(1):108–10.

50. Pornprasertsuk-Damrongsri S, Thanakun S. Carotid artery calcification detected on panoramic radiographs in a group of Thai population. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2006; 101(1):110-5.

51. Khambete N, Kumar R, Risbud M, Joshi A. Reliability of digital panoramic radiographs in detecting calcified carotid artery atheromatous plaques: a clinical study. Indian J Dent Res. 2014; 25(1):36-40.

52. Friedlander A, Maeder L. The prevalence of calcified carotid artery atheromas on the panoramic radiographs of patients with type 2 diabetes mellitus. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 2000; 89(4):420-4.

53. Sisman Y, Ertas E, Gokce C, Menku A, Ulker M, Akgunlu F. The Prevalence of Carotid Artery Calcification on the Panoramic Radiographs in Cappadocia RegionPopulation. Eur J Dent. 2007; 1(3):132-8.

54. Ezoddini-Ardakani F, Mirzaei M, Nayer S, Besharati S, Moeini M. Evaluation of positive predictive value for digital panoramic radiography in comparison to ultrasound in the diagnosis of calcified carotid atheroma. Health. 2014; 6: 1933–40.

55. Romano-Sousa C, Krejci L, Medeiros F, Graciosa-Filho R, Martins M, Guedes V, Fenyo-Pereira M. Diagnostic agreement between panoramic radiographs and color doppler images of carotid atheroma. J Appl Oral Sci. 2009; 17(1): 45-8.

56. Gonçalves J, Yamada J, Berrocal C, Westphalen F, Franco A, Fernandes Â. Prevalence of Pathologic Findings in Panoramic Radiographs: Calcified Carotid Artery Atheroma. Acta Stomatol Croat. 2016; 50(3):230-4.

57. Imanimoghaddam M, Rah Rooh M, Mahmoudi Hashemi E, Javadzade Blouri A. Doppler sonography confirmation in patients showing calcified carotid artery atheroma in panoramic radiography and evaluation of related risk factors. J Dent Res Dent Clin Dent Prospects. 2012; 6(1):6-11.

58. Baryam B, Uckan S, Acikgoz, Müderrisoglu H, Aydinalp A. Digital panoramic radiography: a realiable method to diagnose carotid artery atheromas?. Dentomaxillofacial Radiology. 2006(4); 35: 266-70.

59. North American Symptomatic Carotid Endarterectomy Trial Collaborators. Beneficial effect of carotid endarterectomy in symptomatic patients with high-grade carotid stenosis. N Engl J Med 1991;325:445-53.

60. DeBakey ME. Successful carotid endarterectomy for cerebrovascular insufficiency. Nineteen year follow-up. JAMA 1975; 233: 1083-1085.

61. Barnett HJM, Haines SJ. Carotid endarterectomy for asymptomatic carotid stenosis. New Engl J Med 1993;328:276-278.

ANEXO: A continuación, en el apartado de ´Ánexo´ se exponen la primera página de cada artículo mencionado.

1

Radiopacities in soft tissue on dental radiographs: diagnostic considerations

SADJ March 2015, Vol 70 no 2 p53 - p59

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SUMMARY

Radiopacities in soft tissue in the maxillofacial and oral region frequently manifest on panoramic radiographs in various locations and in several sizes and shapes. Accurate diagnosis is important as the finding may indicate serious disease states. This manuscript provides guidelines for the interpretation of soft tissue radiopacities seen on dental radiographs and recommends additional radiological views required to locate and diagnose the calcifications.

ACRONYMS

CAC: calcified carotid plaque
CBCT: cone beam computed tomography
CTC: calcified triticeous cartilage
GHH: greater horn of hyoid bone
SHTC: superior horn of thyroid cartilage

INTRODUCTION

Soft tissue radiopacities include calcification, ossification or foreign objects. The latter are excluded from this manuscript. Calcification is the deposition of calcium salts in tissue. The pathogenesis is based on either dystrophic or metastatic mechanisms. *Dystrophic calcification*, which comprises the majority of soft tissue calcifications in the head and neck region, is the result of soft tissue damage with tissue degeneration and necrosis which attracts the precipitation of calcium salts. The blood calcium concentration in these patients is normal. Appropriate examples are calcification of a focus of necrosis of tuberculosis, necrotic tumour tissue or of atheromatous plaque.

Metastatic calcification on the other hand results from the deposition of calcium salts in normal tissue in the presence of hypercalcemia secondary to metabolic causes such as hyperparathyroidism and skeletal deposits of malignant disease. Metastatic calcifications are therefore generally spread more widely throughout the body than dystrophic calcifications which tend to be more localized. The radiology literature is ambiguous in distinguishing between soft tissue calcification and ossification as the distinction can often only be made histologically. Soft tissue ossification is the formation of mature bone with or without bone marrow in an extra-skeletal site. Appropriate examples are elongation of the styloid process through ossification of the attached ligaments and bone formation in synovial chondromatosis.

Idiopathic calcification involves normal serum calcium concentration and healthy tissue, and can as such not be classified as either dystrophic or metastatic. Examples of this are tumorous calcinosis which presents with calcifications around joints and calcinosis cutis, which manifests in the cutaneous or subcutaneous tissue overlying the jaw bones. The latter two conditions are rare and will not be discussed further.

Dental practitioners are required to identify, diagnose, treat or refer for treatment all pathology identified on a radiograph. This paper is aimed at providing practitioners with insight into the differential diagnosis of soft tissue radiopacities seen on dental radiographs. In order to achieve this, a thorough knowledge of the anatomic structures in the head and neck area is important. Accurate interpretation relies on correct positioning of the head during radiographic examinations as this may influence the location and visibility of soft tissue radiopacities on the radiograph. Most calcifications require no further management, but there are several which, if not identified and managed appropriately, could have serious health consequences.

PARAMETERS FACILITATING ACCURATE INTERPRETATION

When radiopacities present as an incidental finding in a soft tissue site, it is of pivotal importance to perform a thorough clinical examination which includes history taking and palpation of the respective site. The anatomical position, number of radiopacities, shape- and size of the calcifications and their internal structure provide important guidelines for their accurate interpretation (Table 1).

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Original Article

Prevalence of soft tissue calcifications on digital panoramic radiographs: A retrospective study

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ABSTRACT

Aims and Objectives: To determine the prevalence of visible calcifications in soft tissues of the orofacial region in digital panoramic radiographs. **Materials and Methods:** Panoramic radiographs of 1615 adult male and female dental outpatients who had visited the dental college for various dental treatments were scrutinized for calcifications. Soft tissue calcifications were recorded according to gender, age, and site. **Results:** Patients identified with soft tissue calcifications comprised 63.41% arteriosclerosis, 45.29% calcified atherosclerotic plaques, phlebolith in 11.7%, sialolith of submandibular salivary gland in 4.3%, calcified stylomandibular and stylohyoid ligament in 4.2%, tonsillolith in 3.2% and lymph node calcification in 2.1% of the radiographs. The association of presence of calcification with age was analyzed with the Chi-square test ($P<0.05$). Women showed an increased prevalence of soft tissue calcifications ($P<0.001$). Mean age of participants with calcification and without calcification was assessed ($P<0.05$) using Mann-Whitney *U* test. **Conclusion:** Carotid artery calcifications were found to be high among the soft tissue calcifications and women after menopause showed an increase in the carotid artery calcifications.

Key words: Dystrophic calcification, idiopathic calcification, metastatic calcification, panoramic radiography, prevalence, soft tissue calcifications

Introduction

When calcium salts are deposited in an unorganized fashion in soft tissue, it is called heterotopic calcification, which is divided into three categories: Metastatic, idiopathic and dystrophic. When the serum levels of calcium or phosphate increase, minerals precipitate into normal tissue causing metastatic calcification,[1,2] which usually occurs bilaterally and symmetrically. However, idiopathic calcification occurs in soft tissues even when there are normal serum calcium and phosphate levels. Dystrophic calcification is pathologic and occurs in degenerative and dead tissue despite normal serum calcium and phosphate levels, soft tissue damage caused by trauma, inflammation, injections, presence of parasites, changes arising from disease and calcifications localized to the site of injury.

Calcification of various structures located in the head and neck region are detected accidentally on panoramic radiographs (OPGs) during routine examination of patients seeking dental care.[1] Prevalence of soft

Access this article online

Quick Response Code:

Website: www.jiaomr.in

DOI: 10.4103/0972-1363.155676

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Received: 08-11-2014 **Accepted:** 12-04-2015 **Published:** 22-04-2015

ELSEVIER

Cardiovascular Pathology 13 (2004) 63–70

CARDIOVASCULAR PATHOLOGY

Regulation of cardiovascular calcification

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Received 18 September 2003; received in revised form 27 October 2003; accepted 3 November 2003

Abstract

Vascular calcification is highly correlated with cardiovascular disease (CVD) and is a significant predictor of cardiovascular events, especially in high risk patients such as the end stage renal disease (ESRD) population. Vascular calcification can lead to serious problems including valve stenosis, decreased vascular compliance, calciphylaxis, and even sudden death. However, the contribution of vascular calcification to progression of atherosclerosis is unknown and needs more study. Biochemical, histological, and genetic studies indicate that vascular calcification is actively regulated and involves both positive and negative modulators. Several nonmutually exclusive theories to account for vascular calcification based on current studies are discussed. © 2004 Elsevier Inc. All rights reserved.

Keywords: Modulator; Mutant mouse; Regulation; Vascular calcification

1. Clinical relevance of cardiovascular calcification

Cardiovascular calcification refers to pathological calcium phosphate deposition in the blood vessels, myocardium, and cardiac valves. Clinical consequences of cardiovascular calcification depend on its extent and the organ affected. In the heart, calcification and subsequent stiffening, thickening, and tearing of the valve leaflets have long been known as a major mode of failure of both native as well as bioprosthetic cardiac valves [1,2]. In small arterioles, vascular medial calcification is responsible for calcific uremic arteriolopathy (also called calciphylaxis), an almost-always fatal skin necrotic condition seen in a small but significant percentage of dialysis patients [3]. Vascular medial calcification leading to a stenosing, fibroproliferative arterial process is also the major finding and cause of death in the rare genetic disorder, idiopathic infantile arterial calcification [4].

In contrast to the above observations, calcification of blood vessels commonly seen with aging, uremia, diabetes, and atherosclerosis has been considered, for the past century, a benign finding. This perception is quickly changing as technological advances in noninvasive measurement of vascular calcification, particularly electron beam computed tomography (EBCT), have allowed rapid and sensitive measurements to be correlated to a growing list of clinical events and cardiovascular risk. In coronary arteries, calcification is positively correlated with atherosclerotic plaque burden [5,6], increased risk of myocardial infarction [7–9], and increased risk of dissection following angioplasty [10]. Whether calcification is related to plaque stability, however, is less clear, although recent studies indicate that coronary calcification may be associated with and/or predictive of sudden cardiac death. Using autopsy specimens, intimal calcification was found to be a reliable marker of plaque instability, defined as plaques that have undergone rupture [11]. In addition, in a study of 79 adults with sudden cardiac death, both the Framingham risk index and coronary calcification (as measured by EBCT) were demonstrated to be predictive of future cardiovascular events [12]. Whether these findings relate to increase in plaque instability is controversial; indeed, a recent study using finite element analysis suggested that intimal calcification did not appreciably change the stress profiles of fibrous caps compared to lipid pools [13], though solid shear stresses were not considered thus limiting interpretation of the study.

While some of these findings may relate to the correlation of vascular calcification with extent of underlying arterial disease, it is also possible that vascular calcification itself may contribute to initiation or progression of cardiovascular disease (CVD). This possibility seems particularly plausible in the case of vascular calcification associated with chronic kidney disease (CKD). Over the last 2 years, a

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1054-8807/04/$ – see front matter © 2004 Elsevier Inc. All rights reserved.
doi:10.1016/S1054-8807(03)00130-3

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review

http://www.kidney-international.org

Mechanisms of arterial calcifications and consequences for cardiovascular function

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Cardiovascular complications are the leading cause of mortality in chronic (CKD) and end-stage renal disease (ESRD). The risk of developing cardiovascular complications is associated with changes in the structure and function of the arterial system, which are in many aspects similar to those occurring with aging. The presence of traditional risk factors does not fully explain the extension and severity of arterial disease. Therefore, other factors associated with CKD and ESRD must also be involved. Arterial calcification (AC) is a common complication of CKD and ESRD, and the extent of AC in general population as well as in patients with CKD is predictive of subsequent cardiovascular mortality beyond established conventional risk factors. AC is an active process similar to bone formation that implicates a variety of proteins involved in bone and mineral metabolism and is considered part of a systemic dysfunction defined as CKD-associated mineral and bone disorder (CKD-MBD).

Kidney International Supplements (2013) **3,** 442-445;
doi:10.1038/kisup.2013.92

KEYWORDS: aging; arteriosclerosis; arterial calcifications; arterial stiffness; chronic renal disease

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Premature vascular aging and arterial stiffening are observed with progression of chronic kidney disease (CKD) and in end-stage renal disease (ESRD).[1] Damage of large arteries is a major contributory factor to cardiovascular complications, and a leading cause of mortality in CKD including ESRD.[2,3] Arterial stiffening in such patients is of multifactorial origin, with extensive arterial calcifications (ACs) representing a major covariate.[4–6] ACs are predictive of cardiovascular mortality beyond established conventional risk factors.[7,8] ACs are largely contributing to myocardial ischemia, cardiac hypertrophy, and microvascular disease in brain and kidney.[9,10]

MECHANISMS OF AC

Long time considered a passive process associated with cellular aging and death, and high extracellular fluid calcium–phosphate concentrations, the results of many recent studies have indicated that cardiovascular calcifications are an active process, in many aspects similar to embryonic bone formation, which is regulated by a variety of genes and proteins implicated in mineral and bone metabolism. This process involves differentiation of contractile vascular smooth muscle cells (VSMCs), and pericytes into distinct, 'osteoblast-like' cells with a secretory phenotype. VSMC synthesize bone-associated proteins, including alkaline phosphatase, osteocalcin, osteopontin, and a coat of collagen-rich extracellular matrix, via the formation of matrix vesicles, nodules, and apoptotic bodies, which serve as initiation sites for apatite nanocrystal deposition (but also with bone formation and real ossification).[11–16] Clinical and experimental data show that this process can be triggered by many factors including dyslipidemia,[17] oxidative stress,[18] advanced glycation end-products and hyperglycemia,[19] hyperphosphatemia,[20] increased serum aldosterone levels,[21] and age-associated cellular senescence.[22–24] These risk factors have in common that they all converge to a final pathway, that is, inflammation and nuclear factor (NF)-κB activation.[25–28] Molecular imaging *in vivo* has demonstrated inflammation-associated osteogenesis in early-stages of atherosclerosis,[29] confirming the role of inflammation in triggering the metabolic cascade leading to the transformation of VSMC into an osteogenic phenotype. Macrophage activation releases proinflammatory cytokines (such as interleukin-6 and tumor necrosis factor-α), and proteolytic enzymes (matrix metalloproteinase-2, matrix metalloproteinase-9,

SPECIAL ARTICLE

Re-Evaluation of Risks Associated With Hyperphosphatemia and Hyperparathyroidism in Dialysis Patients: Recommendations for a Change in Management

Geoffrey A. Block, MD, and Friedrich K. Port, MD, MS

• **Hyperphosphatemia is a predictable consequence of chronic renal failure and is present in most patients on dialysis. Traditionally, the risk associated with elevated serum phosphorus has focused on its impact on renal osteodystrophy. A growing body of evidence, however, suggests that abnormalities in serum phosphorus, calcium-phosphorus product (CaxP), and parathyroid hormone (PTH) levels are resulting in vascular and visceral calcification, thereby contributing to the substantially increased risk of cardiovascular death in this population. In this analysis, we review in detail the literature that describes these associations. We show that the current treatment paradigm for serum phosphorus and secondary hyperparathyroidism is ineffective for a large segment of dialysis patients. Currently, 60% of hemodialysis patients have phosphorus greater than 5.5 mg/dL, and 40% have CaxP greater than 60 mg^2/dL^2. It is our belief that prevention of uremic calcification, cardiac death, and vascular disease should assume primary importance when evaluating the risks associated with elevated levels of phosphorus, CaxP, and PTH. We recommend that target levels should become 9.2 to 9.6 mg/dL for calcium, 2.5 to 5.5 mg/dL for phosphorus, less than 55 mg^2/dL^2 for CaxP product, and 100 to 200 pg/mL for intact PTH.**

INDEX WORDS: Phosphate binders; secondary hyperparathyroidism (SHPT); vascular calcification; dialysis; mortality; coronary artery disease; vitamin D.

IT HAS BEEN recognized for nearly three decades that phosphorus (P) is important in the development and progression of secondary hyperparathyroidism.[1] Recently, however, its role in the pathogenesis of this disorder has been greatly clarified. Moreover, serum phosphorus levels have been associated, directly and indirectly, with vascular and visceral calcification. Studies now exist that implicate elevated serum phosphorus, elevated calcium-phosphorus product (CaxP), and secondary hyperparathyroidism (SHPT) in the substantially increased incidence of cardiac, visceral, and peripheral vascular calcification seen in this population. Cardiovascular disease accounts for nearly 50% of all deaths in dialysis patients,[2,3] and the incidence of cardiovascular death is dramatically higher than the cardiovascular mortality seen in the general population (Fig 1).[4] There is growing recognition that abnormal mineral metabolism and secondary hyperparathyroidism together play a key role in the morbidity and mortality of end-stage renal disease (ESRD) patients.

Elevated serum phosphorus is a predictable consequence of ESRD and is present in most patients receiving dialysis. Despite substantial improvements over the last decade in other biochemical parameters, such as Kt/V and hematocrit,[5] little progress has been made in achieving control of serum phosphorus. In 1988, Lowrie and Lew[6] analyzed data from 12,000 hemodialysis patients and found a mean serum phosphorus of 6.2 mg/dL.[6] Our own analysis, using data from 6,400 prevalent hemodialysis patients in 1990 and 1993, again showed a mean phosphorus of 6.2 mg/dL. In the latter study, 39% of patients had a P level greater than 6.5 mg/dL, 30% greater than 7 mg/dL, and 10% greater than 9 mg/dL.[7] Sixty percent of patients had P levels greater than 5.5 mg/dL, the usual upper limit of normal.

Our current strategy for managing hyperphosphatemia and secondary hyperparathyroidism not only has limited effectiveness but also is potentially contributing to cardiovascular morbidity and mortality. We briefly examine the causes for

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Received November 12, 1999; accepted as submitted January 4, 2000.

Supported by an unrestricted educational grant from Genzyme Therapeutics.

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0272-6386/00/3506-0027$3.00/0

doi:10.1053/ajkd.2000.7493

J Am Soc Nephrol 15: 2857–2867, 2004

Human Vascular Smooth Muscle Cells Undergo Vesicle-Mediated Calcification in Response to Changes in Extracellular Calcium and Phosphate Concentrations: A Potential Mechanism for Accelerated Vascular Calcification in ESRD

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Abstract. Patients with ESRD have a high circulating calcium (Ca) × phosphate (P) product and develop extensive vascular calcification that may contribute to their high cardiovascular morbidity. However, the cellular mechanisms underlying vascular calcification in this context are poorly understood. In an *in vitro* model, elevated Ca or P induced human vascular smooth muscle cell (VSMC) calcification independently and synergistically, a process that was potently inhibited by serum. Calcification was initiated by release from living VSMC of membrane-bound matrix vesicles (MV) and also by apoptotic bodies from dying cells. Vesicles released by VSMC after prolonged exposure to Ca and P contained preformed basic calcium phosphate and calcified extensively. However, vesicles released in the presence of serum did not contain basic calcium phosphate, co-purified with the mineralization inhibitor fetuin-A and calcified minimally. Importantly, MV released under normal physiologic conditions did not calcify, and VSMC were also able to inhibit the spontaneous precipitation of Ca and P in solution. The potent mineralization inhibitor matrix Gla protein was found to be present in MV, and pretreatment of VSMC with warfarin markedly enhanced vesicle calcification. These data suggest that in the context of raised Ca and P, vascular calcification is a modifiable, cell-mediated process regulated by vesicle release. These vesicles contain mineralization inhibitors derived from VSMC and serum, and perturbation of the production or function of these inhibitors would lead to accelerated vascular calcification.

Patients with ESRD develop extensive medial calcification, or Monckeberg's sclerosis, that causes increased arterial stiffness and contributes to the high cardiovascular mortality (1,2). Calciphylaxis is an increasingly common and life-threatening form of calcification characterized by destructive calcification in the media of subcutaneous arterioles, leading to occlusion and subsequent widespread tissue necrosis (2,3). The precise pathophysiology of vascular calcification in ESRD is unknown, but risk factors include age, hypertension, time on dialysis, and, most significant, abnormalities in calcium (Ca) and phosphate (P) metabolism (4,5). Normal serum concentrations of Ca and inorganic P ions are metastable with respect to basic calcium phosphate (BCP; a mixture of octacalcium phosphate, dicalcium phosphate dihydrate, and apatite) precipitation but can support growth of nascent crystals. In ESRD, systemic Ca and inorganic P concentrations typically exceed 2.4 and 2.0 mM, respectively (4). Consequently, calcification in ESRD has traditionally been ascribed to supersaturation and subsequent precipitation of mineral ions. This has led to therapeutic measures to reduce the Ca/P product aimed mostly at reduction of P.

Recent studies have shown that vascular calcification is a regulated process similar to bone formation (6,7). VSMC in the normal artery wall constitutively express potent inhibitors of calcification, such as matrix Gla protein (MGP), whose absence results in spontaneous medial calcification (8). In atherosclerotic calcification and diabetic Monckeberg's sclerosis, expression of these endogenous inhibitors is reduced and VSMC express markers of both osteoblast and chondrocyte differentiation (7,9). Moreover, human VSMC in culture spontaneously convert to an osteo/chondrocytic phenotype and

Received March 16, 2004. Accepted June 28, 2004.
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1046-6673/1511-2857
Journal of the American Society of Nephrology

DOI: 10.1097/01.ASN.0000141960.01035.28

Smooth Muscle Cell Phenotypic Transition Associated With Calcification

Upregulation of Cbfa1 and Downregulation of Smooth Muscle Lineage Markers

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Abstract—Bovine aortic smooth muscle cell (BASMC) cultures undergo mineralization on addition of the organic phosphate donor, β-glycerophosphate (βGP). Mineralization is characterized by apatite deposition on collagen fibrils and the presence of matrix vesicles, as has been described in calcified vascular lesions in vivo as well as in bone and teeth. In the present study, we used this model to investigate the molecular mechanisms driving vascular calcification. We found that BASMCs lost their lineage markers, SM22α and smooth muscle α-actin, within 10 days of being placed under calcifying conditions. Conversely, the cells gained an osteogenic phenotype as indicated by an increase in expression and DNA-binding activity of the transcription factor, core binding factor α1 (Cbfa1). Moreover, genes containing the Cbfa1 binding site, OSE2, including osteopontin, osteocalcin, and alkaline phosphatase were elevated. The relevance of these in vitro findings to vascular calcification in vivo was further studied in matrix GLA protein null ($MGP^{-/-}$) mice whose arteries spontaneously calcify. We found that arterial calcification was associated with a similar loss in smooth muscle markers and a gain of osteopontin and Cbfa1 expression. These data demonstrate a novel association of vascular calcification with smooth muscle cell phenotypic transition, in which several osteogenic proteins including osteopontin, osteocalcin, and the bone determining factor Cbfa1 are gained. The findings suggest a positive role for SMCs in promoting vascular calcification. **(*Circ Res*. 2001;89:1147-1154.)**

Key Words: vascular calcification ■ smooth muscle cells ■ phenotype ■ core binding factor α1

Mineralization of bones and teeth is an exquisitely regulated, cell-mediated process in which the tissue extracellular matrix is embedded with crystalline calcium phosphate deposits. This process gives rise to hard tissues endowed with the mechanical properties required to withstand their normal physiological functions. In contrast, mineralization of soft tissues occurs under pathological conditions with detrimental consequences, particularly when present in blood vessels and heart valves. Calcification of arterial plaques decreases vessel elasticity, augments plaque brittleness, leads to increased plaque rupture during angioplasty procedures,[1,2] and is associated with increased risk of myocardial infarction and death.[3,4,5]

Despite its clinical significance, the molecular mechanisms regulating vascular calcification are unclear. Historically, ectopic calcification has been considered a passive process involving spontaneous calcium phosphate mineral precipitation in necrotic tissue. However, several lines of evidence have recently emerged supporting the concept that ectopic calcification, like mineralization of bones and teeth, is a cell-regulated process. Similarities between hard and soft tissue mineralization include the presence of an apatitic mineral phase, matrix vesicles,[6] and noncollagenous bone proteins including osteopontin,[7] bone acidic glycoprotein 75 (BAG 75),[8] osteocalcin,[9] osteonectin,[10] and bone morphogenetic protein type 2.[11] Furthermore, studies of mutant mice, including matrix GLA protein null ($MGP^{-/-}$) mouse,[12] *klotho* mouse,[13] carbonic anhydrase II deficient mouse,[14] and osteoprotegerin null mouse,[15] have identified genes whose loss of function increases susceptibility to vascular calcification. Finally, outright ossification has been occasionally noted in extensively calcified vascular tissues.[16] Taken together, these findings have greatly strengthened the theory that vascular calcification, like osteogenesis, is a delicately regulated balance between inducers and inhibitors.

The importance of vascular calcification in human disease has fueled interest in identifying both positive and negative

Original received March 20, 2000; resubmission received September 27, 2001; revised resubmission received October 19, 2001; accepted October 19, 2001.

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Circulation Research **is available at http://www.circresaha.org** **DOI: 10.1161/hh2401.101070**

© Med Sci Monit, 2012: 18(1): RA1-11
PMID: 22207127

www.MEDSCIMONIT.COM
Review Article

Received: 2010.12.19
Accepted: 2011.05.18
Published: 2012.01.01

The mechanism of vascular calcification – a systematic review

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Source of support: Self financing

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Summary

Calcification of vessels reduces their elasticity, affecting hemodynamic parameters of the cardiovascular system. The development of arterial hypertension, cardiac hypertrophy, ischemic heart disease or peripheral arterial disease significantly increases mortality in patients over 60 years of age. Stage of advancement and the extent of accumulation of calcium deposits in vessel walls are key risk factors of ischemic events.

Vascular calcification is an active and complex process that involves numerous mechanisms responsible for calcium depositions in arterial walls. They lead to increase in arterial stiffness and in pulse wave velocity, which in turn increases cardiovascular disease morbidity and mortality.

In-depth study and thorough understanding of vascular calcification mechanisms may be crucial for establishing an effective vasculoprotective therapy.

The aim of this study was to present a comprehensive survey of current state-of-the-art research into the impact of metabolic and hormonal disorders on development of vascular calcification.

Due to strong resemblance to the processes occurring in bone tissue, drugs used for osteoporosis treatment (calcitriol, estradiol, bisphosphonates) may interfere with the processes occurring in the vessel wall. On the other hand, drugs used to treat cardiovascular problems (statins, angiotensin convertase inhibitors, warfarin, heparins) may have an effect on bone tissue metabolism. Efforts to optimally control calcium and phosphate concentrations are also beneficial for patients with end-stage renal disease, for whom vessel calcification remains a major problem.

key words: **vascular calcification • osteoporosis • menopause • estrogens • raloxifene**

Full-text PDF: http://www.medscimonit.com/fulltxt.php?ICID=882181

Word count: 4838
Tables: 3
Figures: 1
References: 119

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Cell, Vol. 93, 165–176, April 17, 1998, Copyright ©1998 by Cell Press

Osteoprotegerin Ligand Is a Cytokine that Regulates Osteoclast Differentiation and Activation

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Summary

The ligand for osteoprotegerin has been identified, and it is a TNF-related cytokine that replaces the requirement for stromal cells, vitamin D3, and glucocorticoids in the coculture model of in vitro osteoclastogenesis. OPG ligand (OPGL) binds to a unique hematopoeitic progenitor cell that is committed to the osteoclast lineage and stimulates the rapid induction of genes that typify osteoclast development. OPGL directly activates isolated mature osteoclasts in vitro, and short-term administration into normal adult mice results in osteoclast activation associated with systemic hypercalcemia. These data suggest that OPGL is an osteoclast differentiation and activation factor. The effects of OPGL are blocked in vitro and in vivo by OPG, suggesting that OPGL and OPG are key extracellular regulators of osteoclast development.

Introduction

Bone remodeling is a normal process that involves the resorption of bone by osteoclasts and the synthesis of bone matrix by osteoblasts. Osteoclasts are specialized monocyte/macrophage family members that differentiate from hematopoietic precursors (Suda et al., 1992) that appear to stem from a common monocyte precursor. Mature osteoclasts can be formed in vitro from bone marrow and spleen cells, a process that is facilitated by the presence of vitamin D3 and stromal cells (Takahashi et al., 1988). Terminal differentiation in this lineage is characterized by acquisition of mature phenotypic markers, such as the calcitonin receptor, tartrate resistant acid phosphatase (TRAP), integrin $\alpha v\beta 3$, morphological conversion into large multinucleated cells, and the capacity to form resorption lacunae on bone. While indirect evidence suggests that an unidentified osteoclastogenic factor regulates terminal osteoclast development and activation (Horton, 1972; Fuller et al., 1991; Lee et al., 1991; Hentunen et al., 1994), a protein candidate for this factor has not been identified.

Osteoprotegerin (OPG) is a naturally occurring secreted protein with homology to members of the TNF receptor family (Simonet et al., 1997). Administration of OPG in vivo inhibits osteoclastogenesis and associated bone resorption and blocks the pathological increase in osteoclast numbers and activity seen in animal models that mimic osteopenic disorders in humans (Simonet et al., 1997). We hypothesized that OPG might neutralize a factor that stimulates osteoclast development (Simonet et al., 1997), thus inhibiting osteoclast maturation.

Utilizing expression cloning, we have identified a polypeptide ligand for OPG (OPGL), a member of the TNF family of cytokines that exists in transmembrane and soluble (cleaved) forms. OPGL in vitro activates mature osteoclasts and modulates osteoclast formation from bone marrow precursors in the presence of CSF-1 but without the need for vitamin D3 and stromal cells. We demonstrate that it binds to the surface of osteoclast progenitors in CSF-1–treated bone marrow. Recombinant soluble OPGL is a potent inducer of bone resorption in vivo. These data confirm our model in which OPGL and OPG act as positive and negative regulators of osteoclast development.

Results

Identification of OPGL

The biologically active domain of OPG is likely to interact with a TNF family member while negatively regulating osteoclastogenesis in vitro and in vivo. All known members of the TNF family with one exception are type II transmembrane proteins expressed on the cell surface. We used recombinant OPG-Fc fusion protein as an immunoprobe to screen for OPG-binding proteins on the surface of various cell lines and primary hematopoietic cells. The murine myelomonocytic cell line 32D was found to express a surface molecule, which could be detected with both the mouse OPG [22-194]-Fc and human OPG[22-201]-Fc fusion proteins (Figure 1A). Secondary antibody alone did not bind to the surface of 32D cells, nor did purified human IgG1 Fc, indicating that binding of the OPG-Fc fusion proteins was due to the OPG N-terminal moiety. A plasmid cDNA expression library was constructed from 32D cell mRNA and arrayed into pools of 1000 clones. Individual pools were transfected into COS7 cells and then stained with the human OPG [22-201]-Fc fusion protein. One positive pool was identified and subdivided by sequential rounds of sib selection yielding a single plasmid clone, 32D-F3. The 32D-F3 plasmid DNA was then transfected into COS7 cell cultures and then immunostained with either human IgG1 Fc domain, human OPG [22-201]-Fc fusion protein, or with HVEM/ATAR-Fc fusion protein (Hsu et al., 1997), followed by FITC-conjugated secondary antibody. Only

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Osteo/Chondrocytic Transcription Factors and Their Target Genes Exhibit Distinct Patterns of Expression in Human Arterial Calcification

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Objective—Mineralization-regulating proteins are found deposited at sites of vascular calcification. However, the relationship between the onset of calcification in vivo and the expression of genes encoding mineralization-regulating proteins is unknown. This study aimed to determine the temporal and spatial pattern of expression of key bone and cartilage proteins as atherosclerotic calcification progresses.

Methods and Results—Using reverse transcription-polymerase chain reaction on a panel of noncalcified and calcified human arterial samples, two classes of proteins could be identified: (1) Matrix Gla protein, osteonectin, osteoprotegerin, and aggrecan were constitutively expressed by vascular smooth muscle cells (VSMCs) in the normal vessel media but downregulated in calcified arteries whereas (2) alkaline phosphatase, bone sialoprotein, osteocalcin, and collagen II were expressed predominantly in the calcified vessel together with Cbfa1, Msx2, and Sox9, transcription factors that regulate expression of these genes. In the calcified plaque in situ hybridization identified subsets of VSMCs expressing osteoblast and chondrocyte-like gene expression profiles whereas osteoclast-like macrophages were present around sites of calcification.

Conclusions—These observations suggest a sequence of molecular events in vascular calcification beginning with the loss of expression by VSMCs, of constitutive inhibitory proteins, and ending with expression by VSMCs and macrophages of chondrocytic, osteoblastic, and osteoclastic-associated proteins that orchestrate the calcification process. (***Arterioscler Thromb Vasc Biol.*** **2003;23:489-494.)**

Key Words: calcification ■ atherosclerosis ■ osteoblast ■ cartilage ■ vascular smooth muscle cell ■ macrophage

Vascular calcification occurs as a complication of atherosclerosis and involves the nucleation of hydroxyapatite (HA) on membrane-bound vesicles and the local expression/deposition of bone-associated, mineralization-regulating proteins.[1–3] Thus, it shares fundamental similarities with developmental osteogenesis and a feature of many end-stage calcified lesions is the presence of bone trabeculae and/or cartilage-like cells in the vessel wall.[4,5] Until recently little was known of the function of bone-associated proteins in the vasculature, but gene knockout (KO) and in vitro studies have demonstrated that many of them regulate vascular smooth muscle cell (VSMC) phenotype and/or inhibit HA crystal growth.[6–8] Moreover, the vascular phenotypes of the matrix Gla protein (MGP) and osteoprotegerin (OPG) KOs suggest that, in the normal vascular media, calcification is actively inhibited.[8,9]

Studies of human medial calcification (Monckeberg's sclerosis) in diabetes and aging have suggested that the VSMCs that predominate in these lesions lose expression of calcification inhibitors, such as MGP, and begin to express "late" differentiation markers of both osteoblasts (bone sialoprotein; BSP) and osteocalcin (bone Gla protein; BGP) and chondrocytes (collagen II; COLII).[4] This implies that vascular calcification may be caused by phenotypic modulation of resident vascular cells in a permissive matrix environment. However, few studies have explored the phenotype of VSMCs as calcification progresses in atherosclerotic lesions. Bone matrix proteins have been identified at sites of calcification, but it is unclear whether these were expressed locally by VSMCs or deposited from the circulation.[2,3] Moreover, in contrast with Monckeberg's sclerosis, macrophages associate with calcification in atherosclerotic plaques and express the mineralization inhibitor osteopontin, suggesting they may also play a role in regulating mineral deposition.[10]

In vitro studies using both bovine and human VSMCs have shown that VSMCs assume osteo/chondrocytic-like properties whereby they spontaneously, or in response to exogenous factors, co-express numerous osteoblast and chondrocyte markers, form nodules, and calcify over a defined time–course.[4,11] The factors that regulate this phenotypic transition

Received November 13, 2002; revision accepted January 14, 2003.
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Arterioscler Thromb Vasc Biol. **is available at http://www.atvbaha.org** **DOI: 10.1161/01.ATV.0000059406.92165.31**

THE JOURNAL OF BIOLOGICAL CHEMISTRY
Vol. 278, No. 15, Issue of April 11, pp. 13333–13341, 2003
Printed in U.S.A.

Structural Basis of Calcification Inhibition by α_2-HS Glycoprotein/Fetuin-A

FORMATION OF COLLOIDAL CALCIPROTEIN PARTICLES*

Received for publication, October 24, 2002, and in revised form, January 29, 2003
Published, JBC Papers in Press, January 29, 2003, DOI 10.1074/jbc.M210868200

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Genetic evidence from mutant mice suggests that α_2-HS glycoprotein/fetuin-A (Ahsg) is a systemic inhibitor of precipitation of basic calcium phosphate preventing unwanted calcification. Using electron microscopy and dynamic light scattering, we demonstrate that precipitation inhibition by Ahsg is caused by the transient formation of soluble, colloidal spheres, containing Ahsg, calcium, and phosphate. These "calciprotein particles" of 30–150 nm in diameter are initially amorphous and soluble but turn progressively more crystalline and insoluble in a time- and temperature-dependent fashion. Solubilization in Ahsg-containing calciprotein particles provides a novel conceptual framework to explain how insoluble calcium precipitates may be transported and removed in the bodies of mammals. Mutational analysis showed that the basic calcium phosphate precipitation inhibition activity resides in the amino-terminal cystatin-like domain D1 of Ahsg. A structure-function analysis of wild type and mutant forms of cystatin-like domains from Ahsg, full-length fetuin-B, histidine-rich glycoprotein, and kininogen demonstrated that Ahsg domain D1 is most efficient in inhibiting basic calcium phosphate precipitation. The computer-modeled domain structures suggest that a dense array of acidic residues on an extended β-sheet of the cystatin-like domain Ahsg-D1 mediates efficient inhibition.

The combination of mineral with an organic matrix called "biomineral" is commonplace in biology. Biominerals studied in detail include magnetic crystals in bacteria (1), silica skeletons in diatomeous algae (2, 3), shells of marine molluscs (4, 5), and skeletons of vertebrate animals (6). Generally, biominerals form in close proximity with biomacromolecules. Ultrastructural analyses suggest that a protein scaffold provides the ordered and spatially restrained framework for crystal deposition. In mammals, collagen is an excellent scaffold for calcification. Noncollagenous proteins control nucleation, growth, shape, and orientation of crystals in the mineral phase (7, 8). Major mineral ions are equally distributed in the extracellular space of most living organisms. Extracellular fluids are especially supersaturated with regard to calcium and phosphate ions. Therefore, it is surprising that mineralization is restricted to collagenous matrix of the vertebrate skeleton and that once started mineralization does not proceed throughout the organism (9). This suggests that the inhibition of unwanted mineralization is at least as important as the initiation of mineralization. Genetic experimentation with mutant mice indeed suggests that mineralization is the default pathway, which must be actively prevented, not started (10). Unwanted mineralization resulted from the genetic ablation of mineralization inhibitors, pyrophosphate (11, 12) and matrix γ-carboxyl glutamic acid (GLA)[1]-containing protein (MGP) (13). We showed that the lack of α_2-HS glycoprotein/fetuin-A (Ahsg) results in severe systemic calcification in mice and humans (15).[2] Of note, Ahsg is the only protein inhibitor of calcification known so far that is systemic and present throughout the extracellular space in mammals. Due to its high affinity for the mineral phase of bone, Ahsg accumulates about 100-fold over other serum proteins in bones and teeth (16). This seems paradoxical, considering that Ahsg is an efficient inhibitor of calcification both *in vitro* and *in vivo*. Here we studied how Ahsg inhibits the formation of basic calcium phosphate (BCP). Using electron microscopy and dynamic light scattering, we determined that the inhibition is effected by a transient formation of colloidal spheres containing Ahsg, calcium, and phosphate, which we call "calciprotein particles." Further, the structure-function relationship of recombinant forms of Ahsg-like proteins from the cystatin superfamily, fetuin-B (FETUB), histidine-rich glycoprotein (HRG), and kininogen (KNG) suggests that the inhibition of unwanted calcification by Ahsg involves binding of BCP nuclei to an array of acidic amino acid residues on an extended β-sheet of the cystatin-like Ahsg domain D1. We suggest that the resulting diffusion barrier limits further growth of the

* This work was supported by a grant from the Deutsche Forschungsgemeinschaft (to W. J-D.). The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked "*advertisement*" in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

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[1] The abbreviations used are: GLA, γ-carboxyl glutamic acid; MGP, matrix GLA-containing protein; Ahsg, α_2-HS glycoprotein/fetuin-A; BCP, basic calcium phosphate; bAhsg and mAhsg, bovine and mouse Ahsg, respectively; BSA, bovine serum albumin; TEM, transmission electron microscopy; FETUB, fetuin-B; hFETUB and mFETUB, human and mouse FETUB, respectively; HRG, histidine-rich glycoprotein; hHRG, human HRG; KNG, kininogen; hKNG, human KNG; MBP, maltose-binding protein; GST, glutathione S-transferase; HS, α_2-Heremans-Schmid.

[2] C. Schäfer, A. Heiss, A. Schwarz, R. Westenfeld, M. Ketteler, J. Floege, W. Müller-Esterl, T. Schinke, and W. Jahnen-Dechent, submitted for publication.

REVCOG 2009; 14:(3):136-144

Artículo Original

EFECTOS DE LA TERAPIA HORMONAL SUSTITUTIVA Y TIBOLONA SOBRE EL HUESO. Densidad mineral y riesgo de fractura

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Resumen

El tratamiento hormonal (TH) produce incrementos en la densidad mineral ósea en todos los lugares del esqueleto. La reducción del riesgo de fractura ha sido documentada por datos de un metaanálisis, estudios de cohortes y el estudio WHI.
Los estrógenos son una opción terapéutica para la prevención y el tratamiento de la osteoporosis especialmente en la mujer con síntomas climatéricos, con la consideración de que su uso por tiempo prolongado incrementa los riesgos. Por tanto, el tratamiento debe ser individualizado valorando los posibles riesgos personales asociados a la terapia frente a los efectos beneficiosos esperados. Solo así se conseguirá obtener una continuidad en el tratamiento que lleve a conseguir el beneficio buscado en el hueso.
Por su parte, la Tibolona reduce el recambio óseo y mejora de manera significativa la DMO especialmente trabecular. Sin embargo, los estudios incluyen poblaciones pequeñas y tienen una relativamente corta duración (2 años). Los datos sobre fractura vertebral derivan de un único estudio discontinuado precozmente. No hay datos sobre fractura no vertebral.

Abstract

Hormone therapy (HT) during menopause produces increases in bone mineral density in all places of the skeleton. Randomized (including WHI) and cohort studies as well as meta-analysis reports have demonstrated a reduction in the fracture risk.
Estrogens may be considered a therapeutic option for the prevention and treatment of osteoporosis in women with climacteric symptoms keeping in mind that long term use increases the risks of such a therapy. Therefore, treatment should be individualized balancing the potential hazards versus the expected beneficial effects. Thus, only in compliant patients the treatment achieves the benefit sought in bone.
On the other hand, tibolone reduces bone turnover and improve significantly the bone mineral density and quality. However, studies include small populations and have a relatively short duration. Data on vertebral fracture derived from a single study discontinued early and there are no data on non-vertebral fracture.

1. INTRODUCCION

La disminución en la producción de esteroides sexuales por el ovario que ocurre en la peri y postmenopausia, se asocia con una rápida pérdida de masa ósea debida a un incremento en la resorción.
En las dos décadas anteriores, múltiples estudios observacionales han señalado el efecto beneficioso de la terapia hormonal (TH) en la salud de la mujer postmenopáusica, basado fundamentalmente en el alivio de los síntomas asociados a la deprivación estrogénica como síntomas vasomotores y genitourinarios. Estos estudios indicaron además un efecto preventivo de patologías relacionadas con el envejecimiento como la osteoporosis[1].
Los estrógenos han demostrado ser eficaces para incrementar la densidad mineral ósea y prevenir las fracturas, pero la información sobre los efectos secundarios de su uso durante largo tiempo ha reducido su uso para el tratamiento de la osteoporosis .
La tibolona es un esteroide sintético utilizado en el tratamiento de los síntomas climatéricos y disminución de la libido que tiene un efecto agonista estrogénico en el hueso.
Aunque todavía no conocemos totalmente los mecanismos íntimos del control del remodelado óseo, si tenemos información suficiente para afirmar que los estrógenos tienen un papel relevante en la homeostasis del esqueleto y es por ello que su disminución se asocia con reducción en la masa ósea, alteraciones en la microarquitectura y aumento del riesgo de fractura.
En el siguiente capítulo, se analiza el papel de la terapia estrogénica y de la tibolona en la prevención y tratamiento de la osteoporosis postmenopáusica.

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médecine/sciences 2013 ; 29 : 1018-25

Les protéines SIBLING

Outils moléculaires de la progression tumorale et de l'angiogenèse

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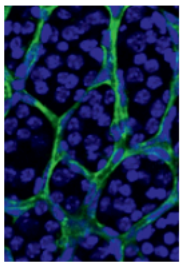
› La famille SIBLING comprend cinq membres : l'ostéopontine (OPN), la sialoprotéine osseuse (BSP), la protéine matricielle de dentine 1 (DMP1), la sialophosphoprotéine de dentine (DSPP) et la phosphoglycoprotéine de matrice extracellulaire (MEPE). Ces protéines ont été regroupées sur la base de leurs caractéristiques biochimiques et génétiques communes. D'abord considérées comme spécifiques des tissus minéralisés de l'os et de la dent, leur surexpression a été ensuite mise en évidence au niveau d'une large variété de tumeurs chez l'homme. Dans cette revue, nous décrivons les rôles attribués aux protéines SIBLING au niveau de chacune des étapes de la progression cancéreuse et métastatique, ainsi que de l'angiogenèse. ‹



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Les protéines de la famille SIBLING

Le regroupement de cinq protéines des matrices minéralisées de l'os et de la dent au sein d'une famille appelée SIBLING (*small integrin-binding ligand N-linkedglycoprotein*) a été proposé pour la première fois par L.W. Fisher *et al.* [1] sur la base de leurs caractéristiques biochimiques et génétiques communes. Les principaux membres de cette famille sont l'ostéopontine (OPN), la sialoprotéine osseuse (BSP), la sialophosphoprotéine de dentine (DSPP), la protéine matricielle de dentine 1 (DMP1) et la phosphoglycoprotéine de matrice extracellulaire (MEPE). Ces protéines, hautement glycosylées et phosphorylées, ont d'abord été isolées à partir des matrices extracellulaires minéralisées des os et de la dent. Alors que l'OPN est la protéine SIBLING la mieux étudiée, les autres protéines ont connu un regain d'intérêt ces 20 dernières années, surtout après la mise en évidence de leur expression ectopique en dehors des tissus minéralisés et, notamment, au niveau des cellules tumorales malignes. L'objet de cette revue est de décrire les propriétés des protéines SIBLING dans le contexte du cancer. Ainsi, leur expression importante par différents types de cancers chez l'homme (pour une revue détaillée, voir [2]) ne sera mentionnée ici que dans quelques exemples utiles.

Chez l'homme, les gènes codant pour les SIBLING sont localisés au niveau d'une région de 375 000 paires de bases sur le bras long du chromosome 4. L.W. Fisher et *al.* ont observé que ces gènes présentent de nombreuses similitudes, notamment au niveau de l'agencement de leurs exons *(Figure 1)*, suggérant qu'ils proviendraient d'un même gène ancestral ayant subi des duplications et des modifications au cours du temps [1].

Les cinq protéines partagent plusieurs domaines fonctionnels, tels que des sites de liaison au calcium, le motif RGD (Arg-Glyc-Asp) qui assure l'interaction entre ces protéines et les récepteurs de surface cellulaire de type intégrine, et le domaine appelé ASARM (*acidic serine and aspartate-rich motif*) qui est impliqué dans la minéralisation osseuse *(Figure 1)*. D'autres domaines sont plus spécifiques de certaines SIBLING comme la séquence cryptique SVVYGLR *(Figure 1)* qui permet la liaison de l'OPN aux intégrines $\alpha_9\beta_1$ et $\alpha_4\beta_1$. De manière générale, ces protéines lient les récepteurs de type intégrine et le récepteur CD44[1] *(Tableau I)*. Ces interactions activent des voies de signalisation aboutissant à l'adhésion, la migration et la survie cellulaires.

Les activités biologiques des SIBLING sont modulées par des processus protéolytiques qui peuvent révéler des sites de liaison cryptiques et/ou supprimer des domaines fonctionnels influençant, notamment, l'adhésion et la migration cellulaires. Les fonctions connues des SIBLING au

Vignette (Photo © Inserm - Nicolas Ricard et Didier Grunwald).

[1] CD44 désigne une famille de protéines résultant de l'épissage alternatif de 10 exons du gène, fonctionnant dans la réponse immune et comme récepteur de l'acide hyaluronique [51].

m/s n° 11, vol. 29, novembre 2013
DOI : 10.1051/medsci/20132911019

Angiología. 2016;68(6):459–464

Angiología

www.elsevier.es/angiologia

ORIGINAL

La osteopontina como biomarcador de riesgo neurológico en la enfermedad carotídea

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Recibido el 26 de abril de 2016; aceptado el 1 de junio de 2016
Disponible en Internet el 1 de julio de 2016

PALABRAS CLAVE
Osteopontina;
Estenosis carotídea;
Placa inestable

Resumen

Introducción: La osteopontina (OPN) incrementa el reclutamiento, migración y adhesión de los macrófagos y modula la expresión de citocinas proinflamatorias e interleucinas. Actualmente, no está definida su asociación con la inestabilidad de la placa de ateroma carotídea y la sintomatología clínica de los pacientes.

Objetivos: Estudiar los niveles en plasma de OPN de pacientes intervenidos quirúrgicamente de endarterectomía carotídea (ECA) y correlacionarlos con la sintomatología clínica preoperatoria, con el fin de valorar el riesgo neurológico y la inestabilidad de placa.

Material y métodos: Se diseñó un estudio prospectivo con una muestra de pacientes consecutivos intervenidos quirúrgicamente de ECA, previamente evaluados por el neurólogo o con la realización de una TAC o RMN cerebral. Los pacientes se dividieron en 2 grupos (sintomáticos y asintomáticos) y se compararon con un grupo control. Se excluyeron aquellos con enfermedades intercurrentes. La OPN se determinó mediante enzimoinmunoanálisis. Se utilizó para el análisis estadístico el programa SPSS v. 18.0. Las variables categóricas se describen como frecuencias y las cuantitativas como media y desviación estándar en el caso de utilizar pruebas paramétricas, y como mediana y rango intercuartil en el caso de utilizar pruebas no paramétricas. Se estableció que la relación fue estadísticamente significativa si p era inferior a 0,05.

Resultados: Durante el periodo de estudio, 44 pacientes (39 hombres, 5 mujeres), de edad media 75 ± 6,62 años, fueron intervenidos de ECA por presentar una placa de ateroma que producía una estenosis significativa (>70% con ecodoppler). De acuerdo con sus antecedentes cerebrovasculares, 24 fueron sintomáticos y 20 asintomáticos. El grupo control fue de 25 sujetos sanos. La OPN en el grupo control fue de 60 ± 6,62 ng/mL, de 74,3 ± 60,8 ng/mL en asintomáticos y de 90,3 ± 45,4 ng/mL en sintomáticos (p = 0,003).

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http://dx.doi.org/10.1016/j.angio.2016.06.001

Osteopontin: A Multifunctional Protein at the Crossroads of Inflammation, Atherosclerosis, and Vascular Calcification

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Current Atherosclerosis Reports 2009, **11**:206–213
Current Medicine Group LLC ISSN 1523-3804

Osteopontin (OPN) was initially identified in osteoblasts as a mineralization-modulatory matrix protein. Recently, OPN has been studied as a multifunctional protein that is upregulated in a variety of acute and chronic inflammatory conditions, such as wound healing, fibrosis, autoimmune disease, and atherosclerosis. OPN is highly expressed at sites with atherosclerotic plaques, especially those associated with macrophages and foam cells. In the context of atherosclerosis, OPN is generally regarded as a proinflammatory and proatherogenic molecule. However, the role of OPN in vascular calcification (VC), which is closely related to chronic and active inflammation, is that of a negative regulator because it is an inhibitor of calcification and an active inducer of decalcification. OPN expression and its regulatory molecular mechanisms remain elusive during the process of VC. Therefore, further research with regard to the role of OPN in diseases associated with VC is needed to identify potential OPN-related therapeutic targets.

Introduction

Osteopontin (OPN) is a protein that belongs to the small intertrin-binding N-linked glycoprotein (SIBLING) family. This family possesses an arginine-glycine-aspartate (RGD) binding site and includes bone sialoprotein, dentin matrix protein 1, dentin sialophosphoprotein, and matrix extracellular phosphoglycoprotein [1]. OPN was first identified in 1986 in an osteosarcoma as a matrix protein with the potential to bridge cells and hydroxyapatite. OPN is also an extracellular structural protein and an organic component of bone [2]. Bone matrix consists of about 90% type I collagen and about 5% noncollagenous proteins. These noncollagenous proteins include bone sialoprotein, thrombospondin, fibronectin, vitronectin, and OPN, all of which contain RGD sequences that interact with some integrins [3]. OPN has several cell adhesive domains; these domain structures contain 13 phosphorylation sites. The RGD-containing domains interact with cell surface integrins (eg, αvβ3, αvβ1, and αvβ5). OPN is also referred to as a secreted phosphoprotein 1, which is a major phosphoprotein secreted in cultured cells, or as an early T-cell activation factor, which is a cytokine produced by activated lymphocytes and macrophages [4]. In this aspect, OPN regulates recruitment of inflammatory cells and adhesion or migration of tumor cells by binding to integrins (αvβ3, αvβ1, αvβ5, and α4β1) or the splice variant of CD44 v3-v6, which is an OPN receptor [5]. An increased OPN level may be altered by extracellular enzymes containing thrombin and kinases [6]. This article reviews the role of OPN in a variety of inflammatory conditions and focuses especially on atherosclerosis and vascular calcification (VC).

Cellular Sources

OPN is expressed and secreted in various tissue and cells, including brain, liver, the gastrointestinal tract, the lungs, bone, cardiac tissues, joints, and kidneys. In addition, it is found in a variety of biological fluids, including blood, urine, milk, and seminal fluid. In normal tissue, OPN is expressed in preosteoblasts, osteoblasts, osteocytes, smooth muscle cells, and endothelial cells (ECs) [6]. During wound healing or under proinflammatory conditions, OPN expression is specifically increased near inflammatory cells [7]. OPN is produced as a cytokine in activated T cells and macrophages, suggesting that OPN plays an important role in the modulation of inflammation [8]. In clinical studies of patients with chronic inflammation, autoimmune disease,

REVIEWS

J Am Soc Nephrol 15: 2959–2964, 2004

Vascular Calcification Mechanisms

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Abstract. Vascular calcification is highly correlated with cardiovascular disease mortality, especially in patients with ESRD or diabetes. In addition to the devastating effects of inappropriate biomineralization seen in cardiac valvulopathies, calciphylaxis, and idiopathic arterial calcification, vascular calcification is now recognized as a marker of atherosclerotic plaque burden as well as a major contributor to loss of arterial compliance and increased pulse pressure seen with age, diabetes, and renal insufficiency. In recent years, several mechanisms to explain vascular calcification have been identified including (*1*) loss of inhibition, (*2*) induction of bone formation, (*3*) circulating nucleational complexes, and (*4*) cell death. Alterations in calcium (Ca) and phosphorus (P) balance as seen in patients with ESRD promotes vascular calcification via multiple mechanisms and may explain the alarmingly high levels of cardiovascular disease deaths in these patients. Strategies to control Ca and P levels in patients with ESRD have met with early success in preventing progression of vascular calcification. Whether or not vascular calcification can be reversed is not yet known, but exciting new studies suggest that this may be possible in the future.

Pathologic calcification of cardiovascular structures, or vascular calcification, is associated with a number of diseases including ESRD and cardiovascular disease. Calcium phosphate deposition, in the form of bioapatite, is the hallmark of vascular calcification and can occur in the blood vessels, myocardium, and cardiac valves. In blood vessels, calcified deposits are found in distinct layers of the blood vessel and are related to underlying pathology. Intimal calcification occurs in atherosclerotic lesions (1,2), whereas medial calcification (also known as Monckeberg's medial sclerosis) is associated with vascular stiffening and arteriosclerosis observed with age, diabetes, and ESRD (3,4). Intimal calcification may occur independently of medial calcification and *vice versa*. In patients with ESRD, a mixture of intimal and medial calcification has been observed in affected vessels (5,6).

Clinical Consequences of Vascular Calcification

Vascular calcification can lead to devastating organ dysfunction depending on its extent and the organ affected. In the heart, calcification of cardiac valve leaflets is recognized as a major mode of failure of native as well as bioprosthetic valves (7,8). In dialysis patients, vascular medial calcification is responsible for calcific uremic arteriolopathy, a necrotizing skin condition associated with extremely high mortality rates (9). Finally, a genetic deficiency in pyrophosphate levels causes idiopathic infantile arterial calcification, a disease characterized by arterial calcification, fibrosis, and stenosis that leads to premature death in affected neonates (10).

In contrast, calcification of blood vessels commonly seen with aging, ESRD, diabetes, and atherosclerosis has historically been considered a benign finding. However, the introduction of new techniques to measure vascular calcification noninvasively, such as electron beam computed tomography, have revolutionized our current thinking about the risks of vascular calcification. In coronary arteries, calcification is positively correlated with atherosclerotic plaque burden (11,12), increased risk of myocardial infarction (13–15), and plaque instability (2,16). Although some of these findings may relate to the correlation of coronary calcification with extent of underlying atherosclerotic disease, it is also possible that vascular calcification itself may contribute to initiation or progression of cardiovascular disease (CVD). This possibility seems particularly plausible in the case of coronary calcification associated with ESRD (see below). Finally, vascular calcification, especially that found in the media of large arteries, leads to increased stiffening and therefore decreased compliance of these vessels. The consequent loss of the important cushioning function of these arteries is associated with increased arterial pulse wave velocity and pulse pressure, and leads to impaired arterial distensibility, increased afterload favoring left ventricular hypertrophy, and compromised coronary perfusion (17,18). Indeed, medial arterial calcification is strongly correlated with coronary artery disease and future cardiovascular events in patients with type 1 (19,20) and is a strong prognostic marker of CVD mortality in patients with ESRD (21). Thus, vascular calcification has a profound influence on cardiovascular function and health.

Cardiovascular Calcification and CVD Mortality in ESRD

More than half the deaths in patients with ESRD are due to CVD. In fact, the risk of CVD mortality in adult patients with ESRD is 20 to 30 times higher than that of the general popu-

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1046-6673/1512-2959
Journal of the American Society of Nephrology

DOI: 10.1097/01.ASN.0000145894.57533.C4

Kidney International, Vol. 63 (2003), pp. 1003–1011

Uremia induces the osteoblast differentiation factor Cbfa1 in human blood vessels

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Uremia induces the osteoblast differentiation factor Cbfa1 in human blood vessels.

Background. Bone matrix proteins are expressed in calcified arteries from dialysis patients, suggesting that vascular smooth muscle cells (VSMCs) may transform to osteoblast-like cells. One of the key transcriptional regulators of osteoblast differentiation is Cbfa1. Thus, we hypothesized that this may be a key factor in arterial calcification.

Methods. To test this hypothesis, we examined sections of the inferior epigastric artery from uremic patients for the presence of Cbfa1 and type I collagen and osteopontin by in situ hybridization and immunostaining. We also examined the effect of pooled uremic sera from dialysis patients on the expression of Cbfa1 by reverse transcription-polymerase chain reaction (RT-PCR) in bovine VSMCs in vitro.

Results. Cbfa1 and osteopontin were expressed in both the media and the intima in vessels that were calcified, but there was only minimal staining in non-calcified vessels. In vitro studies demonstrated that pooled uremic serum, compared to pooled control human serum induced the expression of Cbfa1 by RT-PCR in bovine VSMCs in a time-dependent, nonphosphorus-mediated mechanism.

Conclusion. These results support that Cbfa1 is a key regulatory factor in the vascular calcification observed in dialysis patients and is up-regulated in response to many uremic toxins.

The assessment of coronary arteries by new noninvasive imaging techniques such as electron beam computed tomography scan (EBCT) and intracardiac ultrasound has heightened the awareness that over 70% of atherosclerotic plaques observed in the aging population are calcified [1], and the magnitude of calcification correlates with the severity of obstructive coronary artery disease by angiography and clinical events [2, 3]. Cardiovascular disease and stroke are the leading causes of death in patients with end-stage renal disease (ESRD) that require dialysis, at a risk that is 10- to 20-fold greater than the general population [4, 5]. Studies evaluating coronary calcification by EBCT in patients with ESRD have demonstrated excessive coronary artery calcification, even in young adults [6, 7]. Pathologic analysis of arteries from nondialysis patients demonstrates that vascular calcification resembles developmental bone mineralization, with the production of "bone" proteins by vascular smooth muscle cells (VSMCs), such as osteopontin, bone sialoprotein, alkaline phosphatase, and type I collagen [8–11]. We have confirmed these findings in arteries from dialysis patients [12, 13]. This would imply that the calcification of vascular tissue is an active process, with VSMCs transforming to osteoblast-like cells. However, the mechanism by which this occurs is not yet clear.

In vitro experiments in both human and bovine VSMCs have demonstrated that phosphorus, in the form of β-glycerophosphate (which is cleaved by alkaline phosphatase to form free phosphate), can induce calcification similar to that observed in osteoblast cultures [14, 15]. The mechanism by which phosphorus induced calcification was dependent on the sodium-phosphate (Na/Pi) co-transporter [16]. Furthermore, in a recent study by Jono et al [17], exogenous phosphate added to human VSMCs culture up-regulated Cbfa1 expression. Cbfa1 is a transcription factor critical for osteoblast differentiation and the expression of the bone matrix proteins, osteopontin, osteocalcin, and type I collagen [18]. In addition, Cbfa1 knockout mice fail to form mineralized bone, proving that Cbfa1 is critical for the terminal differentiation of osteoblasts [19]. Thus, the in vitro data in VSMCs support that phosphorus can lead to calcification, and that phosphorus can induce Cbfa1 and the expression of bone matrix proteins. However, in vivo evidence of this relationship to vascular calcification is lacking. We have previously demonstrated that pooled serum collected from hemodialysis patients can also induce mineralization of bovine VSMCs to a greater extent and at an earlier time point than bovine VSMCs incubated with pooled serum from healthy controls [20]. In addition,

Key words: Cbfa1, vascular calcification, osteopontin, type 1 collagen, dialysis.

Received for publication August 16, 2002
and in revised form October 4, 2002
Accepted for publication October 17, 2002

Molecular Medicine

Induction of Bone-Type Alkaline Phosphatase in Human Vascular Smooth Muscle Cells

Roles of Tumor Necrosis Factor-α and Oncostatin M Derived From Macrophages

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Abstract—Inflammatory cells such as macrophages and T lymphocytes play an important role in vascular calcification associated with atherosclerosis and cardiac valvular disease. In particular, macrophages activated with cytokines derived from T lymphocytes such as interferon-γ (IFN-γ) may contribute to the development of vascular calcification. Moreover, we have shown the stimulatory effect of 1α,25-dihydroxyvitamin D_3 (1,25$(OH)_2D_3$) on in vitro calcification through increasing the expression of alkaline phosphatase (ALP), an ectoenzyme indispensable for bone mineralization, in vascular smooth muscle cells. Therefore, we hypothesized that macrophages may induce calcifying phenotype, especially the expression of ALP in human vascular smooth muscle cells (HVSMCs) in the presence of IFN-γ and 1,25$(OH)_2D_3$. To test this hypothesis, we used cocultures of HVSMCs with human monocytic cell line (THP-1) or peripheral blood monocytes (PBMCs) in the presence of IFN-γ and 1,25$(OH)_2D_3$. THP-1 cells or PBMCs induced ALP activity and its gene expression in HVSMCs and the cells with high expression of ALP calcified their extracellular matrix by the addition of β-glycerophosphate. Thermostability and immunoassay showed that ALP induced in HVSMCs was bone-specific enzyme. We further identified tumor necrosis factor-α (TNF-α) and oncostatin M (OSM) as major factors inducing ALP in HVSMCs in the culture supernatants of THP-1 cells. TNF-α and OSM, only when applied together, increased ALP activities and in vitro calcification in HVSMCs in the presence of IFN-γ and 1,25$(OH)_2D_3$. These results suggest that macrophages may contribute to the development of vascular calcification through producing various inflammatory mediators, especially TNF-α and OSM. **(*Circ Res*. 2002;91:9-16.)**

Key Words: vascular calcification ■ alkaline phosphatase ■ macrophages ■ tumor necrosis factor-α ■ oncostatin M

Dystrophic calcification is often associated with inflammatory vascular diseases such as atherosclerosis and cardiac valvular disease.[1–5] Inflammatory cells such as macrophages and T lymphocytes are frequently found in advanced atherosclerotic lesions, particularly fibrofatty or fibrous plaques in which calcification usually initiates.[6,7] Calcium deposits are usually observed at the periphery of the lipid core. It has been pointed out that macrophages are the predominant cell type associated with different stages of calcification in atherosclerotic plaques.[8] T-lymphocyte and macrophage infiltrates are also associated with calcification in human native and porcine bioprosthetic valves, and it has been suggested that production of noncollagenous proteins and cytokines by these cells may contribute to valvular calcification.[9,10] Therefore, T lymphocytes and macrophages may play an important role in the development of vascular calcification.

Vascular cells such as vascular smooth muscle cells (VSMCs), pericyte-like cells, and valvular cells play an important role in vascular calcification. VSMCs and pericyte-like cells derived from bovine aorta have calcifying capacity and express noncollagenous matrix proteins such as osteopontin (OPN), matrix gla protein (MGP), and osteocalcin (OC).[11–14] VSMCs also produce osteoprotegerin (OPG), which is a soluble decoy receptor of the tumor necrosis factor receptor superfamily and may regulate vascular calcification as well as skeletal metabolism.[15] Valvular cells derived from aortic valves can also calcify their extracellular matrix.[16] In vitro calcification by vascular cells can be modulated by calciotropic hormones, steroid hormones, transforming growth factor-β, and 25-hydroxycholesterol.[13,16–18] These results suggest that phenotypic changes of vascular cells in mesenchymal origin, especially acquisition of calcifying phenotype under various pathological conditions, may contribute to the development of vascular calcification.

Original received January 15, 2002; revision received June 5, 2002; accepted June 5, 2002.
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Circulation Research **is available at http://www.circresaha.org** **DOI: 10.1161/01.RES.0000026421.61398.F2**

Growth Factors, December 2004 Vol. 22 (4), pp. 233–241

Taylor & Francis healthsciences

Bone Morphogenetic Proteins

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(Received 19 March 2004; Revised 17 June 2004)

Bone morphogenetic proteins (BMPs) are multi-functional growth factors that belong to the transforming growth factor β (TGFβ) superfamily. The roles of BMPs in embryonic development and cellular functions in postnatal and adult animals have been extensively studied in recent years. Signal transduction studies have revealed that Smad1, 5 and 8 are the immediate downstream molecules of BMP receptors and play a central role in BMP signal transduction. Studies from transgenic and knockout mice and from animals and humans with naturally occurring mutations in BMPs and related genes have shown that BMP signaling plays critical roles in heart, neural and cartilage development. BMPs also play an important role in postnatal bone formation. BMP activities are regulated at different molecular levels. Preclinical and clinical studies have shown that BMP-2 can be utilized in various therapeutic interventions such as bone defects, non-union fractures, spinal fusion, osteoporosis and root canal surgery. Tissue-specific knockout of a specific BMP ligand, a subtype of BMP receptors or a specific signaling molecule is required to further determine the specific role of a BMP ligand, receptor or signaling molecule in a particular tissue.

BMPs are members of the TGFβ superfamily. The activity of BMPs was first identified in the 1960s (Urist, M.R. (1965) "Bone formation by autoinduction", *Science* **150**, 893–899), but the proteins responsible for bone induction remained unknown until the purification and sequence of bovine BMP-3 (osteogenin) and cloning of human BMP-2 and 4 in the late 1980s (Wozney, J.M. *et al.* (1988) "Novel regulators of bone formation: molecular clones and activities", *Science* **242**, 1528–1534; Luyten, F.P. *et al.* (1989) "Purification and partial amino acid sequence of osteogenin, a protein initiating bone differentiation", *J. Biol. Chem.* **264**, 13377–13380; Wozney, J.M. (1992) "The bone morphogenetic protein family and osteogenesis", *Mol. Reprod. Dev.* **32**, 160–167). To date, around 20 BMP family members have been identified and characterized. BMPs signal through serine/threonine kinase receptors, composed of type I and II subtypes. Three type I receptors have been shown to bind BMP ligands, type IA and IB BMP receptors (BMPR-IA or ALK-3 and BMPR-IB or ALK-6) and type IA activin receptor (ActR-IA or ALK-2) (Koenig, B.B. *et al.* (1994) "Characterization and cloning of a receptor for BMP-2 and BMP-4 from NIH 3T3 cells", *Mol. Cell. Biol.* **14**, 5961–5974; ten Dijke, P. *et al.* (1994) "Identification of type I receptors for osteogenic protein-1 and bone morphogenetic protein-4", *J. Biol. Chem.* **269**, 16985–16988; Macias-Silva, M. *et al.* (1998) "Specific activation of Smad1 signaling pathways by the BMP7 type I receptor, ALK2", *J. Biol. Chem.* **273**, 25628–25636). Three type II receptors for BMPs have also been identified and they are type II BMP receptor (BMPR-II) and type II and IIB activin receptors (ActR-II and ActR-IIB) (Yamashita, H. *et al.* (1995) "Osteogenic protein-1 binds to activin type II receptors and induces certain activin-like effects", *J. Cell. Biol.* **130**, 217–226; Rosenzweig, B.L. *et al.* (1995) "Cloning and characterization of a human type II receptor for bone morphogenetic proteins", *Proc. Natl Acad. Sci. USA* **92**, 7632–7636; Kawabata, M. *et al.* (1995) "Cloning of a novel type II serine/threonine kinase receptor through interaction with the type I transforming growth factor-β receptor", *J. Biol. Chem.* **270**, 5625–5630). Whereas BMPR-IA, IB and II are specific to BMPs, ActR-IA, II and IIB are also signaling receptors for activins. These receptors are expressed differentially in various tissues. Type I and II BMP receptors are both indispensable for signal transduction. After ligand binding they form a heterotetrameric-activated receptor complex consisting of two pairs of a type I and II receptor complex (Moustakas, A. and C.H. Heldi (2002) "From mono- to oligo-Smads: the heart of the matter in TGFβ signal transduction" *Genes Dev.* **16**, 67–871). The type I BMP receptor substrates include a protein family, the Smad proteins, that play a central role in relaying the BMP signal from the receptor to target genes in the nucleus. Smad1, 5 and 8 are phosphorylated by BMP receptors in a ligand-dependent manner (Hoodless, P.A. *et al.* (1996) "MADR1, a MAD-related protein that functions in BMP2 signaling pathways", *Cell* **85**, 489–500; Chen Y. *et al.* (1997) "Smad8 mediates the signaling of the receptor serine kinase", *Proc. Natl Acad. Sci. USA* **94**, 12938–12943; Nishimura R. *et al.* (1998) "Smad5 and DPC4 are key molecules in

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ISSN 0897-7194 print/ISSN 1029-2292 online © 2004 Taylor & Francis Ltd
DOI: 10.1080/08977190412331279890

Cell, Vol. 89, 747–754, May 30, 1997, Copyright ©1997 by Cell Press

Osf2/Cbfa1: A Transcriptional Activator of Osteoblast Differentiation

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Summary

The osteoblast is the bone-forming cell. The molecular basis of osteoblast-specific gene expression and differentiation is unknown. We previously identified an osteoblast-specific cis-acting element, termed OSE2, in the Osteocalcin promoter. We have now cloned the cDNA encoding Osf2/Cbfa1, the protein that binds to OSE2. Osf2/Cbfa1 expression is initiated in the mesenchymal condensations of the developing skeleton, is strictly restricted to cells of the osteoblast lineage thereafter, and is regulated by BMP7 and vitamin D_3. Osf2/Cbfa1 binds to and regulates the expression of multiple genes expressed in osteoblasts. Finally, forced expression of Osf2/Cbfa1 in nonosteoblastic cells induces the expression of the principal osteoblast-specific genes. This study identifies Osf2/Cbfa1 as an osteoblast-specific transcription factor and as a regulator of osteoblast differentiation.

Introduction

Skeletal development is a multistep process. It includes patterning of skeletal elements, commitment of mesenchymal cells to chondrogenic and osteogenic lineages, and terminal differentiation of precursor cells into three specialized cell types: the chondrocyte in cartilage and the osteoblast and osteoclast in bone. Many genes encoding either growth factors or transcription factors were shown through genetic studies to control skeleton patterning (for review, see Cohn and Tickle, 1996; Hogan, 1996). These genetic analyses also showed that mutations in these genes do not severely affect the differentiation of the skeleton-specific cell types, suggesting that patterning and cell differentiation in the skeleton are achieved through different genetic pathways. Consistent with this hypothesis, genes such as PTHrP and c-fos were shown to control chondrocyte and osteoclast differentiation, respectively, without affecting skeletal patterning (Johnson et al., 1992; Wang et al., 1992; Karaplis et al., 1994).

The osteoblast is a cell of mesenchymal origin that, once terminally differentiated, produces most of the proteins present in the bone extracellular matrix (ECM) and controls the mineralization of this ECM. As such, it is viewed as the bone-forming cell. Progress in understanding osteoblast differentiation has been hampered by the small number of molecular markers truly specific to the osteoblast and by the absence of a morphologic feature distinguishing this cell from a fibroblast (Aubin and Liu, 1996).

It is likely that differentiation along the osteoblast lineage involves osteoblast-specific transcription factors (OSFs) that have yet to be identified. To search for OSFs, we and others studied the regulation of expression of Osteocalcin, the only gene expressed in osteoblasts and in no other ECM-producing cell type (Towler et al., 1994; Ducy and Karsenty, 1995). We initially characterized a cis-acting element, termed OSE2, in the promoter of the mouse Osteocalcin gene 2 (OG2) that binds a factor present only in osteoblasts nuclear extracts and confers osteoblast-specific activity on a heterologous promoter (Ducy and Karsenty, 1995). Analysis of Osf2, the osteoblast nuclear activity binding to OSE2, showed that it is immunologically related to the Cbfa transcription factors (Geoffroy et al., 1995; Merriman et al., 1995). The Cbfa proteins are the mouse homologs of Runt, a Drosophila pair-rule gene product (Gergen and Wieschaus, 1985). Runt and the Cbfa proteins have a high degree of homology in their DNA-binding domain, a 128 amino acid-long motif called the runt domain (Kagoshima et al., 1993). The mouse genome contains three known runt homologs encoding numerous isoforms with well-characterized expression patterns (Bae et al., 1992; Ogawa et al., 1993; Wijmenga et al., 1995; Simeone et al., 1995). None of the described Cbfa transcripts has been shown to be expressed predominantly in bone, suggesting that a still unknown member(s) of the Cbfa family controls osteoblast-specific expression of Osteocalcin.

In this paper, we report the cloning of Osf2/Cbfa1, the factor that binds to the OSE2 element. Osf2/Cbfa1 has several functional features that identify it as a transcriptional regulator of osteoblast differentiation.

Results

Isolation and Expression of Osf2/Cbfa1

To search for a Cbfa-related mRNA in osteoblasts, we performed Northern blot analysis using poly(A)$^+$ RNA from mouse thymus and spleen, two tissues expressing the known Cbfas, and from primary osteoblasts. Hybridization with a probe containing sequences coding for the runt domain of Cbfa1, a gene thought to be expressed in T lymphocytes (Satake et al., 1995), detected a transcript in osteoblasts that was at least 20-fold more abundant than the signal detected in thymus (Figure 1A). Thus, we screened at reduced stringency a mouse osteoblast cDNA library using this probe. Three independent clones encoding the same protein were identified. This cDNA was called Osf2/Cbfa1 because it encodes the factor binding to OSE2 and is encoded by the Cbfa1 gene (see below).

Osf2/Cbfa1 contains a glutamine/alanine-rich domain at its N-terminal end, a runt domain, and a proline/serine/

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Proc. Natl. Acad. Sci. USA
Vol. 87, pp. 2220–2224, March 1990
Biochemistry

Recombinant human bone morphogenetic protein induces bone formation

(cartilage induction)

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Communicated by Tom Maniatis, December 14, 1989

ABSTRACT **We have purified and characterized active recombinant human bone morphogenetic protein (BMP) 2A. Implantation of the recombinant protein in rats showed that a single BMP can induce bone formation *in vivo*. A dose–response and time-course study using the rat ectopic bone formation assay revealed that implantation of 0.5–115 μg of partially purified recombinant human BMP-2A resulted in cartilage by day 7 and bone formation by day 14. The time at which bone formation occurred was dependent on the amount of BMP-2A implanted; at high doses bone formation could be observed at 5 days. The cartilage- and bone-inductive activity of the recombinant BMP-2A is histologically indistinguishable from that of bone extracts. Thus, recombinant BMP-2A has therapeutic potential to promote *de novo* bone formation in humans.**

The therapeutic potential for bone formation induced by demineralized bone or its extracts has long been recognized (1–4), but the definition of the factor(s) responsible has remained elusive. We previously described the molecular cloning of the genes for bone morphogenetic protein (BMP) 1, 2A, 2B, and 3, using peptide sequence information from a group of proteins purified from such an extract (5, 6). Each of these proteins was implicated in cartilage and bone formation by preliminary experiments which demonstrated *in vivo* cartilage induction at 7 days (5) in the rat ectopic bone-formation system (7). We now describe the purification and characterization of recombinant human BMP-2A, produced by a Chinese hamster ovary (CHO) cell line, and its activity in ectopic bone formation.

METHODS

Purification. CHO cells (line 2ΔD) were grown in Dulbecco's modified Eagle's medium (DMEM)/Ham's nutrient mixture F-12, 1:1 (vol/vol), supplemented with 10% fetal bovine serum. When the cells were 80–100% confluent, the medium was replaced with serum-free DMEM/F-12; medium was harvested every 24 hr for 4 days. Thirty-seven liters of conditioned medium was directly applied to an 80-ml heparin-Sepharose (Pharmacia) column. The resin was washed with 0.15 M NaCl/6 M urea/20 mM Tris, pH 7.4, and then developed with a linear gradient to 1 M NaCl/6 M urea/50 mM Tris, pH 7.4. Fractions were assayed for *in vivo* cartilage and bone formation after reconstitution of protein with a collagenous matrix (6, 7). The fractions with highest specific activity were pooled and concentrated by ultrafiltration with a YM10 membrane (Amicon). Conditioned medium from CHO cells not transfected with the BMP-2A gene was prepared similarly, except that a step gradient to 1 M NaCl was used.

Further purification was achieved by preparative $NaDodSO_4$/PAGE (8). Approximately 300 μg of protein was applied to a 1.5-mm-thick 12.5% gel; recovery was estimated by adding L-[^{35}S]methionine-labeled BMP-2A, purified over heparin-Sepharose as above. Protein was visualized by copper staining of an adjacent lane (9), appropriate bands were excised and extracted in 0.1% $NaDodSO_4$/20 mM Tris, pH 8.0, and then proteins were desalted on a 5.0 × 0.46-cm Vydac C_4 column (The Separations Group, Hesperia, CA) in 0.1% trifluoroacetic acid in acetonitrile (6).

Protein concentration was determined by amino acid analysis.

Immunological Methods. A fragment of BMP-2A (amino acids 130–396) produced in inclusion bodies in *Escherichia coli* (provided by John McCoy, Genetics Institute) was purified by $NaDodSO_4$/PAGE under reducing conditions (8), eluted from the gel, and used to immunize rabbits (antibody 130). Peptides (Applied Biosystems) were conjugated to thyroglobulin or bovine serum albumin with glutaraldehyde (10) and used to immunize turkeys. Animals were initially injected with 500 μg of protein mixed with complete Freund's adjuvant and then given biweekly booster injections with 250–125 μg of protein in incomplete Freund's adjuvant. Immunoblots (11) reacted with rabbit antiserum were visualized with ^{125}I-labeled protein A (New England Nuclear); immunoblots were incubated with turkey antisera in the presence of thyroglobulin or bovine serum albumin (100 μg/ml) and then visualized with ^{125}I-labeled rabbit anti-turkey IgG (12).

RESULTS

To achieve high levels of BMP-2A protein expression, the gene for BMP-2A was inserted into a mammalian expression vector, stably introduced into CHO cells, and amplified to high copy number by methotrexate selection of dihydrofolate reductase (13). Individual cell lines were selected for study after preliminary examination of levels of BMP-2A mRNA. Production of BMP-2A was analyzed by using antisera prepared against denatured BMP-2A produced in *E. coli* (referred to as antibody 130, made against a 30-kDa C-terminal fragment; Fig. 1*C*), a peptide of amino acids 103–115 (antibody 103, N-terminal region), or a peptide of amino acids 350–365 (antibody 350, C-terminal region). Direct assay of conditioned medium in the *in vivo* cartilage and bone induction assay was not reliable, probably because of the exogenous proteins present even in serum-free medium. The major secreted proteins in the conditioned medium from one CHO

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Abbreviation: BMP, bone morphogenetic protein.
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showed no response to naloxone. Three months later he was admitted in respiratory failure refractory to all treatment. Necropsy showed chronic bronchitis, bullous emphysema, and right ventricular hypertrophy. The intercostal muscles were thin and fibrous, and histology of these and the diaphragm and psoas showed changes of chronic denervation. Histology of the spinal cord was not obtained.

Discussion

Naloxone reverses the apnoeic response to hypoxia in neonatal rabbits[2] but, even in large doses, has no effect on respiration in hypoxic man.[3] Respiratory failure in this patient was thought to be caused by a combination of chronic airflow obstruction and weakness of the respiratory muscles (possibly due to chronic spinal muscular atrophy).[4] Intravenous naloxone produced an increase in minute ventilation and general agitation when he was acutely ill. The increase in oxygen saturation was greater than expected for the increase in ventilation, suggesting improved ventilation-perfusion matching, but the response occurred only in the acute illness.

These findings suggest that there may be overproduction of, or increased sensitivity to, endorphins in acute respiratory failure. Naloxone is beneficial in shock,[5] the postulated mechanism being that endorphins inhibit the interaction of catecholamines with their receptors. Such an action might account for the changes in ventilation-perfusion balance in our patient.

Requests for reprints should be sent to Dr J Ayres, Department of Thoracic Medicine, Guy's Hospital, London SE1 9RT.

References

1 Santiago TV, Remolina C, Scoles V, Edelman NH. Endorphins and the control of breathing. *N Engl J Med* 1981;**304**:1190-5.
2 Grunstein MM, Hazinski TA, Schlueter MA. Respiratory control during hypoxia in newborn rabbits: implied action of endorphins. *J Appl Physiol* 1981;**51**:122-30.
3 Fleetham JA, Clarke H, Dhingra S, Chernick V, Anthonisen NR. Endogenous opiates and chemical control of breathing in humans. *Am Rev Respir Dis* 1980;**121**:1045-9.
4 Haas H, Johnson LR, Gill TH, Armentrout TS. Diaphragm paralysis and ventilatory failure in chronic proximal spinal muscular atrophy. *Am Rev Respir Dis* 1981;**123**:465-76.
5 Anonymous. Naloxone for septic shock. *Lancet* 1981;i:538-9.

(*Accepted 8 January 1982*)

Medial arterial calcification and diabetic neuropathy

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Abstract

X-ray examinations of the feet, knees, and hands were performed on 20 diabetics with severe neuropathy and 20 diabetics with no evidence of neuropathy but with a similar mean age and duration of diabetes. All were under 53 years old with no clinical evidence of peripheral vascular disease. Medial arterial calcification was much more common and extensive in the patients with neuropathy, occurring in the feet in 15 and in the hands in eight compared with in four ($p < 0.001$) and none ($p < 0.001$) of the controls respectively. Although there was some correlation between calcification and both proteinuria ($p < 0.05$) and proliferative retinopathy ($p < 0.02$), the association between calcification and neuropathy ($p < 0.001$) was much stronger.

Neuropathy, with sympathetic denervation of the smooth muscle of the tunica media, may be important in the aetiology of medial arterial calcification.

Introduction

Medial arterial calcification, otherwise known as Monckeberg's sclerosis,[1] was described in diabetics in 1924,[2] but its aetiology and importance remain unknown. It is easily detected on *x*-ray films by its classical "pipe-stem" or "tramline" appearance (see fig 1). Previous observations have indicated that the calcification is predominantly related to the age of the patient and the duration of diabetes.[3-5] During a recent study of blood flow in the foot in patients with neuropathy, however, it was noted that medial wall calcification was particularly common in diabetics with severe neuropathy.[6] There were two aims of the present study: firstly, to determine whether medial wall calcification was a specific complication of diabetes, not just related to age and duration of disease, and, secondly, to consider its relation to neuropathy, which could be important in its pathogenesis.

Patients and methods

The presence of calcification was determined in two groups of patients, the first consisting of 20 diabetics with neuropathy and the

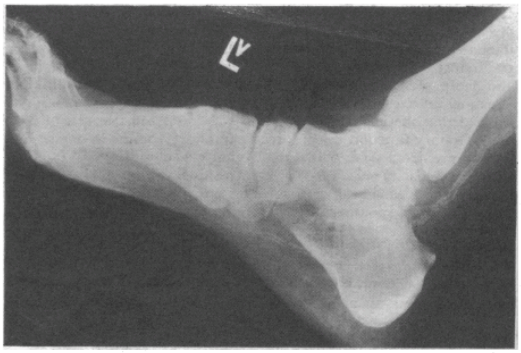

FIG 1—Lateral view of foot showing characteristic "tramline" appearance of medial arterial calcification.

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Calcificación vascular: tipos y mecanismos

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Nefrologia 2011;31(2):142-7

doi:10.3265/Nefrologia.pre2010.Nov.10754

RESUMEN

Clásicamente se consideraba que la calcificación vascular era un proceso pasivo y degenerativo que frecuentemente ocurría con la edad avanzada, aterosclerosis, varias alteraciones metabólicas (como diabetes mellitus y estadios finales de enfermedad renal) y en raras enfermedades genéticas. Sin embargo, desde hace algunos años, la calcificación vascular es considerada como un proceso activo y regulado de manera semejante a la mineralización y metabolismo del hueso, en el que se encuentran implicadas diversas proteínas óseas. Resultados recientes cuestionan la clásica separación de la calcificación vascular en calcificación de la íntima y calcificación de la media, al menos en arterias de capacitancia. Mecanismos procalcificantes y anticalcificantes desempeñan un papel activo en la deposición de calcio en las células vasculares, por lo que su estudio se ha convertido en un área muy activa de investigación. La identificación de dianas terapéuticas que puedan enlentecer o incluso revertir la calcificación vascular podría suponer un avance muy importante en las estrategias terapéuticas para los pacientes afectados de enfermedades renales.

Palabras clave: Calcificación de la media. Calcificación de la íntima. Enfermedad renal crónica. Calcificación vascular.

Vascular calcification: types and mechanisms

ABSTRACT

Vascular calcification has traditionally been considered to be a passive process that was associated with advanced age, atherosclerosis, uncommon genetic diseases and some metabolic alterations such as diabetes mellitus and end-stage kidney failure. However, in the last years, vascular calcification has been proven to be an active and regulated process, similar to bone mineralisation, in which different bone-related proteins are involved. Recent results question the classic classification of vascular calcification into intimal and medial calcification, at least in capacitance arteries. Pro and anti-calcifying mechanisms play an active role in calcium deposition in vascular cells, making this area an active focus of research. The identification of therapeutic targets which can slow down the progression or even reverse vascular calcification could be an important step forward in the treatment of patients with chronic kidney disease.

Keywords: *Medial calcification. Intimal calcification. End-stage kidney disease. Vascular calcification.*

INTRODUCCIÓN

En la calcificación vascular la deposición de fosfato cálcico, en forma de cristales de bioapatita (similar al hueso), puede ocurrir en los vasos sanguíneos y en las válvulas cardíacas[1]. Clásicamente, se han distinguido los tipos de clacificación arterial dependiendo de dónde se depositara el calcio. Así, la calcificación arterial se ha dividido en calcificación de la íntima (asociada a la placa de ateroma)[2], y en calcificación de la media (conocida como esclerosis de Mönckeberg), ligada a la rigidez vascular por mineralización de las fibras elásticas y la arteriosclerosis observada con la edad, diabetes y enfermedad renal crónica (ERC)[3]. La primera estaría relacionada con un aumento de la deposición de lípidos y el infiltrado de células inflamatorias mientras que en la segunda tendría más influencia el cambio de fenotipo de las células de músculo

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European Heart Journal Advance Access published April 16, 2014

European Heart Journal
doi:10.1093/eurheartj/ehu163

REVIEW

Clinical update

Medial vascular calcification revisited: review and perspectives

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Received 28 September 2013; revised 6 March 2014; accepted 21 March 2014

Vascular calcifications (VCs) are actively regulated biological processes associated with crystallization of hydroxyapatite in the extracellular matrix and in cells of the *media* (VCm) or *intima* (VCi) of the arterial wall. Both patterns of VC often coincide and occur in patients with type II diabetes, chronic kidney disease, and other less frequent disorders; VCs are also typical in senile degeneration. In this article, we review the current state of knowledge about the pathology, molecular biology, and nosology of VCm, expand on potential mechanisms responsible for poor prognosis, and expose some of the directions for future research in this area.

Keywords Vascular calcifications • Mönckeberg's media sclerosis • Vascular molecular biology and genetics • Vascular function

Introduction

Vascular calcifications (VCs) are of similar composition to bone minerals. Vascular calcification of the media (VCm) are, in principal, deposits of hydroxyapatite with a high degree of crystallization.[1] Initially, VCs were thought to be the result of passive degenerative processes; however, recent studies illustrate that VC is an active process initiated and regulated via a variety of molecular signalling pathways.[2] While considerable progress elucidating the signalling pathways regulating VC formation has been achieved, the exact molecular basis of VC still remains elusive.[3]

With incoming new research data, the already large number of molecular mechanisms suggested to contribute to VC formation continues to grow. It appears that while deposition of hydroxyapatite represents the resulting commonality of VC, different initiating and propagating molecular mechanisms, as well as diverse crystalline compositions of calcium apatite crystals may be present in various forms of VC.[4–6] For example, it seems likely that VC processes associated with atheroma formation may be triggered by specific biochemical cascades that are altogether different from the cascades initiated by primary damage to elastic fibres which occurs in medial calcification; however, both ultimately result in ectopic VC. While experimental conditions in a given model may replicate parts of the calcification process, they may not provide the whole picture in any of these specific conditions.

We believe that some of the emerging molecular complexity of VC may be accounted for by differences in experimental designs, tendencies to unite a host of different findings associated with different types of VC under a single umbrella hypothesis, undue focus on specific molecular cascades replicating parts of the calcification process in a given model, as well as disregard for specific aetiologies of VC types.

In this article, we review the current state of knowledge about the clinical pathology, molecular biology, and nosology of VCm and expose potential directions for future research in this exciting and clinically important area.

Nomenclature

Establishing common nomenclature is not only the first step to sharing wisdom but also critical to focus research directions and interpret the results. While molecular pathogenesis-based nomenclature of VC is not available, provisory descriptive terminology should supply the need at present (*Figure 1*). While VCi is commonly associated with atherosclerotic plaques, VCm represents a group of

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Evaluation of calcified carotid artery atheromas detected by panoramic radiograph among 80-year-olds

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Objective. To evaluate the incidence among 80-year-olds of calcified carotid artery atheromas (CCAAs) as detected on panoramic radiographs. The relationship between CCAAs and general and oral health was also evaluated.
Study design. Six hundred and fifty-nine panoramic radiographs (262 males, 397 females), obtained from 80-year-old residents of Fukuoka Prefecture, Japan, were used for evaluation of CCAAs.
Results. Of 659 panoramic radiographs, 33 (5%) were noted to have CCAAs. These appeared as a radiopaque nodular mass or masses adjacent to or just below the intervertebral space between C3 and C4. CCAAs were found in 8 males and 25 females. There were marginally significant differences between males and females in CCAAs ($P = 0.06$). Seventy-four percent of CCAAs were detected in the right side. There appeared to be very little relationship between CCAAs and general and oral health.
Conclusions. The results of this study gives further support to the idea of using panoramic radiographs to detect CCAAs. Therefore, we feel that panoramic radiographs should be evaluated not only for pathosis of the teeth and jaws, but also for other incidental findings, especially in the soft-tissue region of the neck. The findings from this study provide potentially life-saving information especially for those elderly people who are at risk for stroke.
(Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2003;96:647-50)

With the number of aged persons increasing worldwide, the number of oral and other diseases will increase exponentially. As early as 1998, the Japanese Ministry of Health and Welfare conceived of a project evaluating the relationship between oral health and general health in those individuals who were 80 years old in Fukuoka Prefecture. The study in this paper makes extensive use of panoramic radiographs obtained during this project.

One of the results obtained from this project, as reported by Ansai et al,[1] was that a higher proportion of those 80-year-olds who were physically active retained a higher number of their teeth and had a higher bone density. From the same project, Takata et al[2] noted that tooth loss was an independent predictor of abnormal electrocardiograph findings, including ST depression, T-wave abnormalities, and arrhythmias.

Friedlander and Lande[3] studied the detection of calcified carotid artery plaque (calcified carotid artery atheroma, or CCAA) by means of panoramic radiography. CCAAs are projected on panoramic radiographs as a radiopaque nodular mass or masses adjacent to the cervical vertebrae at or below the intervertebral space between C3 and C4.[4]

Since their study, similar research using panoramic radiographs has been done.[4-8] From these studies it appears that panoramic radiography is a useful imaging modality for the identification of some asymptomatic patients at the high end of risk for stroke.[5] Using this method, a training program for detection of CCAAs on panoramic radiographs was recently reported by Almog et al.[8] In that report a differential diagnosis of CCAAs included a detailed description of anatomic and pathologic radiopacity.

The purpose of our study was to evaluate the incidence of CCAAs detected on panoramic radiographs of this 80-year-old patient population. Further, we evaluated the relationship between CCAAs and general and oral health examinations.

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1079-2104/2003/$30.00 + 0
doi:10.1016/j.tripleo.2003.07.001

Evaluation of a training program for detection of carotid artery calcifications on panoramic radiographs

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Objective. To evaluate the effectiveness of the American Academy of Oral and Maxillofacial Radiology-sponsored training packet for identification of carotid artery calcifications on panoramic radiographs.
Study design. Two examiners, who completed the training (trainees), examined 778 panoramic radiographs. The sample included 298 men, with a mean age of 66, and 480 women, with a mean age of 68. Findings were compared with those obtained by an oral and maxillofacial radiologist. A kappa statistic was used to determine agreement between the 2 trainees. The positive predictive value (PPV) of the program was estimated by comparing the trainees rating of disease status with an expert in case identification.
Results. Examiners 1 and 2 identified 99 and 78 positive cases, respectively. A kappa statistic of 0.87 (95% CI, 0.81-0.92) was obtained, indicating good interexaminer agreement. The expert identified 27 positive cases, resulting in a PPV of 34.6% (95% CI, 24.4-46.3).
Conclusion. Although the training packet offers valuable training, it does not provide a high PPV, suggesting the need to modify it or to seek an expert opinion before classification of a patient as having calcification on a panoramic radiograph.
(Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2000;90:111-7)

Stroke or cerebrovascular accident is the third leading cause of death in the United States today, preceded only by cardiovascular disease and cancer. It is also the leading cause of severe disability. Each year, about 12 of every 10,000 Americans have a stroke, and there are currently 3.8 million people who have had strokes who are alive.[1] People over 55 have a greater risk of having a stroke, and this risk increases as one gets older.

There are 2 main types of strokes, ischemic and hemorrhagic. About 80% of strokes are ischemic and are caused by atherosclerosis (ie, a buildup of cholesterol-containing fatty deposits called plaque) that roughens the arterial endothelium. This irregular surface can cause turbulent blood flow around the buildup and can trigger development of a clot. More than half of ischemic strokes are caused by stationary (thrombotic) blood clots that develop in the carotid arteries. Temporary, and usually brief, symptoms stemming from the disruption in blood supply describe a transient ischemic attack (TIA). During a TIA, the body may release enzymes that dissolve the clot quickly and restore the blood flow. The remainder of the ischemic strokes are caused by stenosis, largely in the area of the carotid bifurcation. Hemorrhagic strokes occur when a blood vessel in the brain leaks or ruptures. Blood from the hemorrhage spills into the surrounding brain tissue, causing damage. Brain cells beyond the leak or rupture are deprived of blood and are also damaged.[1]

From a clinical perspective, carotid stenosis after a Doppler ultrasound study is typically categorized as one of the following: (A) normal: 0% stenosis, (B) mild: 1% to 39% diameter reduction, (C) moderate: 40% to 59% diameter reduction, (D) severe: 60% to 79% diameter reduction, (E) critical: 80% to 99% diameter reduction, and (F) occluded: 100% diameter reduction.[2-5]

To determine appropriate treatment for carotid artery stenosis, a number of randomized controlled clinical trials took place between 1991 and 1995, including the North American Symptomatic Carotid Endarterectomy Trial (NASCET),[6] the European Carotid Surgery Trial,[7] the Asymptomatic Carotid Atherosclerosis Study (ACAS),[8] and the Efficacy of Carotid Endarterectomy for Asymptomatic Carotid Stenosis study.[9] These studies have helped to define the role of

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Received for publication Nov 10, 1999; accepted for publication Jan 30, 2000.

1079-2104/2000/$12.00 + 0 **7/16/107056**
doi:10.1067/moe.2000.107056

Short Communications

Rupture of Atheromatous Plaque as a Cause of Thrombotic Occlusion of Stenotic Internal Carotid Artery

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We analyzed the clinical profiles and autopsy findings of five patients who died shortly after developing cerebral infarction following thrombotic occlusion of the internal carotid artery. In all five cases, thrombotic occlusion was caused by rupture of the fibrous lining over the gruel of atheroma at the origin of the internal carotid artery showing tight stenosis of the lumen. The mean±SD shorter diameter of the lumen at the site of occlusion was 1.5±0.4 mm. Our results show that an internal carotid artery with tight stenosis of the lumen by atheroma containing gruel harbors a risk of thrombotic occlusion, which may give rise to cerebral infarction by artery-to-artery embolism or by reduced cerebral perfusion. (*Stroke* 1990;21:1740–1745)

Although plaque morphology of patients with atherosclerotic disease of the internal carotid artery (ICA) has been evaluated by clinical means such as angiography, B-mode scanning, and Doppler ultrasonography and by histologic examination of carotid endarterectomy specimens,[1] there are only a few reports analyzing mechanisms of thrombotic occlusion of the ICA in patients who died shortly after the occlusion.[2,3] Postmortem study, which enables us to examine the entire length of the extracranial and intracranial cervicocephalic arteries and the brain, is a golden opportunity to analyze plaque complication leading to thrombotic occlusion of the ICA and the mechanism for occurrence of stroke. We had previously reported a patient with recent thrombotic occlusion of a stenotic ICA due to rupture of the plaque.[2] We extensively reviewed all the autopsy cases in our hospital and found four more patients with recent thrombotic occlusion of the ICA, which urged us to report the clinical features and postmortem pathologic findings of the five patients to add to the body of knowledge that would influence the formulation of a treatment plan.

Subjects and Methods

Among 1,550 patients autopsied in the National Cardiovascular Center from 1977 to 1988, ICA occlusion was found in 48. Of the 48 patients, five died ≤60 days after developing thrombotic occlusion of an atherosclerotic ICA, 16 (including one with a recent thrombus on the other side [case 2 in this report]) had an atherosclerotic ICA occluded by organized thrombi, 26 had an ICA occluded by fresh or organized thromboemboli of cardiac origin, and two (one suffering from Behçet's syndrome and the other from aortitis syndrome) had an ICA occluded by organized thrombi. We analyzed the clinical profiles and autopsy findings of the five patients with recent thrombotic occlusion of the ICA. These patients comprised three men and two women aged 61–78 (mean 69) years (Table 1).

At autopsy, the brain and extracranial arteries were removed and fixed in 10% formalin. After fixation, the arteries at the base of the brain and the extracranial arteries were decalcified in 45% formic acid in 10% sodium citrate solution for 2 days, then histologically examined at approximately 3-mm intervals with hematoxylin and eosin staining and other methods as needed.

The clinical and pathologic features of case 5 have been reported previously.[2]

Results

Table 1 summarizes the profiles of the five patients. All were hypertensive, and case 1 had diabetes mellitus. Case 2 experienced right hemiparesis 7 years before the last stroke and dysarthria lasting for several days 10 months before the last stroke, case 4 experienced a single transient ischemic attack consisting of left hemiparesis 9 days before the last stroke, and case 5 developed cerebral infarction due to occlusion of a cortical branch of the left middle

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Received July 7, 1989; accepted July 24, 1990.

Utility of panoramic radiographs in detecting cervical calcified carotid atheroma

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Objective. The objective of this study was to determine the utility of panoramic radiographs for detecting extracranial calcified carotid atheroma and carotid luminal stenosis.
Study design. Panoramic radiographs were obtained on 52 adult participants who had carotid ultrasound examination. Extent of carotid calcification and stenosis was determined by a cardiologist from ultrasound reports, which were considered gold standard assessments. A trained and calibrated oral and maxillofacial radiologist interpreted the radiographs for presence or absence of carotid calcifications. We examined the utility of panoramic radiographs to diagnose any carotid artery changes (diagnostic scheme 1) or only moderate to severe changes (scheme 2). Generalized estimating equations were used to account for clustering of observations within subjects.
Results. Under diagnostic schemes 1 and 2, radiographs had low sensitivity to detect carotid calcifications (31.1% and 25.0%, respectively) and stenoses (22.7% and 21.4%, respectively).
Conclusions. When compared to ultrasonography, panoramic radiography is not a reliable means to detect carotid artery calcifications or stenoses. **(Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2007;103:543-8)**

Stroke, or cerebrovascular accident, is the third leading cause of death and a major cause of long-term disability in the United States.[1] The estimated health care cost of stroke in the United States for 2006 is $57.9 billion.[1] Arterial stenosis caused by carotid atheromas is one of the many risk factors for stroke. Carotid atheroma is an atherosclerotic disease process that occurs along the walls of the lumen of the common carotid artery near its bifurcation. Pieces of the atheroma may ulcerate and break off to form an embolus that can occlude a smaller intracerebral artery causing stroke.

Several studies suggest that panoramic radiographs can be used to detect calcified carotid atheromas.[2-5] Calcifications in the carotid bifurcation region are detectable on panoramic radiographs in 1% to 5% of the adult population.[4,6] Such findings are more prevalent in certain groups, such as patients who have had renal transplantation and dialysis,[7] patients with type 2 diabetes mellitus,[8] and patients who have had head and neck radiation therapy.[9] Current recommendations suggest that patients with calcified carotid atheromas diagnosed via panoramic radiography should be referred to their physician for cardiovascular assessment.[6,10-12] Few studies, however, have determined the utility of this diagnostic approach.[13]

For the extracranial carotid vasculature, ultrasonography is considered the "gold standard" of noninvasive techniques to determine calcification and stenosis caused by atherosclerotic disease.[14] Identifying the level of stenosis or degree of lumen obstruction is critical as it is correlated with possibility of future stroke.[14-17] To our knowledge, no studies have determined the utility of panoramic radiographs for detecting both calcified carotid atheroma *and* stenosis. The purpose of this study was to determine the utility of panoramic radiographs for detecting extracranial calcified carotid atheroma and carotid luminal stenosis as diagnosed by carotid ultrasonography.

SUBJECTS AND METHODS

Subjects

Patients who had recently undergone carotid ultrasonography for any reason were recruited for this study.

This study was supported by Oral Health Clinical Research Center, University of Minnesota School of Dentistry, and HealthPartners Research Foundation, Minneapolis, MN.
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Received for publication Apr 19, 2006; returned for revision Jun 9, 2006; accepted for publication Jun 14, 2006.
1079-2104/$ - see front matter

doi:10.1016/j.tripleo.2006.06.048

RESEARCH
radiology

Screening panoramic radiology of adults in general dental practice: radiological findings

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Aim To identify the radiological findings from routine screening panoramic radiographs taken of adult (≥ 18 years) patients in general dental practice.
Method Forty-one general dental practitioners (GDPs) who routinely took panoramic radiographs of all new adult patients were recruited. In total, they submitted 1,818 panoramic radiographs of consecutive patients along with basic patient information, radiological reports and treatment plans. The radiographs were also reported by 'experts' (consensus of two dental radiologists). Radiological findings were recorded from the GDP assessments (dentist RY), the experts (expert RY), after exclusion of findings that would have been seen on posterior bitewing radiographs (MRY) and after exclusion of findings of no relevance to treatment (MRYT).
Results There was no significant difference in age profile between the study sample and Dental Practice Board population figures ($P = 0.26$). No radiographs other than the panoramic radiograph had been taken for 57.1% of patients. For the GDP assessments, only 4.6% of patients had radiographs with no radiological findings, while for the experts this proportion was 3.1%. With the exception of the assessment of periodontal bone loss, the experts diagnosed significantly greater proportions of cases as having positive radiological findings. Agreement between dentist and expert assessments varied greatly. When findings from bitewing radiographs were excluded, no radiological findings were recorded on the radiographs of 17.2% of patients. When proposed treatment plans were taken into account, the majority of patients' radiographs (56.3%) had no radiological findings of relevance to treatment.
Conclusions The choice of radiographic examination for the majority of patients in the study did not follow current guidelines. Dentists diagnosed fewer abnormalities than did experts. While many radiological findings are revealed by panoramic radiography, these may either duplicate information from bitewing radiographs or are often of no significance to treatment planning. This study did not provide evidence to support the practice of routine panoramic radiography of all new adult patients.

The proliferation of panoramic radiography in general dental practice in the United Kingdom over the last two decades has been remarkable, rising from 0.7 million in 1981 to 2.04 million in 1998/9.[1] In 1994, the National Radiological Protection Board estimated that there were 3,250 panoramic x-ray sets in use in the United Kingdom.[2]

It is a fundamental requirement of radiation protection that all exposures to x-rays as part of diagnosis should be clinically justified for each patient.[3] Nevertheless, a recent questionnaire study[4] found that 42% of dentists with panoramic x-ray equipment carried out routine panoramic radiography of all new adult patients. This practice of 'screening' has been condemned in recent evidence-based guidelines.[5] In order to justify routine panoramic screening, it would be necessary to demonstrate a significant diagnostic yield that outweighed the risks of the x-ray exposure.

A number of studies, reviewed by Rushton and Horner, have measured the diagnostic yield obtained from panoramic radiology.[6] However, these studies were not performed in general dental practice, instead they were surveys of hospital patients or of specific target groups of individuals. Furthermore, these previous studies have recorded all radiological findings, regardless of whether they were of clinical significance or whether they would have been identified on posterior bitewing radiography. The latter is recommended[5] as an essential aid to diagnosis in initial examination of dentate patients and, in such instances, panoramic radiography may simply duplicate diagnostic yield already available to the dentist.

The aim of this study was to identify the radiological findings from routine screening panoramic radiographs taken of adult (18 years and over) patients in general dental practice, taking into account the findings that would have been identified on posterior bitewing radiographs and the relevance to treatment of the findings.

Materials and methods

In 1997, a questionnaire-based study was performed which addressed various aspects of panoramic radiography in general dental practice. The study, previously reported,[4,7] received completed questionnaires from 542 dentists (73.3% response rate). Forty-two per cent of respondents identified themselves as always taking a panoramic radiograph of new adult (≥ 18 years) patients. To these practitioners, a letter was sent inviting them to participate in a prospective study on panoramic radiography.

A total of 41 dentists agreed to take part in the study, which commenced in the autumn of 1998 and concluded in the late winter of 1999. Each dentist was asked to provide, prospectively, the panoramic radiographs of 50 consecutive new adult (>18 years) patients. In addition, the dentists were asked to include a radiological report ('dentist assessment') for the panoramic radiograph of each patient. Details of the checklist given to the dentists for the purposes of the radiological report are given in Table 1. The radiological findings from the panoramic radiographs is subsequently described here as the Radiological Yield (RY).

Finally, the dentists were required to provide basic patient information and give details of their proposed treatment plan for each patient. Each plan was submitted to the investigators, along with the

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REFEREED PAPER
Received 05.06.00; Accepted 09.11.00
© *British Dental Journal* 2001; **190**: 495–501

J Oral Maxillofac Surg
52:1257-1262, 1994

Panoramic Dental Radiography as an Aid in Detecting Patients at Risk for Stroke

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Purpose: Atherosclerotic lesions in the region of the bifurcation of the common carotid artery and in the internal carotid artery are the most common cause of stroke. On occasion these lesions are calcified and visible on a panoramic dental radiograph.

Methods: Six subjects receiving outpatient dental treatment and denying a history of previous transient ischemic attacks or stroke had bilateral calcified carotid arterial lesions noted on their routine panoramic dental radiograph.

Results: Electronic thermography (ET) demonstrated that these patients had significant temperature differences bilaterally between their medial supraorbital region and the ipsilateral remainder of their forehead when compared with control subjects. These findings are consistent with the presence of calcified stenotic intraluminal plaques altering blood flow, tissue perfusion, and skin temperature readings. The presence of stenotic plaques was verified by Doppler spectral analysis and imaging.

Conclusion: ET of the face, currently considered an investigational procedure, demonstrates promise as an ancillary imaging system capable of confirming the diagnosis of patients at risk of stroke. Such individuals should be referred to an appropriate physician for consideration of medications and/or surgical removal of the plaque. In selected individuals, these are safe and relatively reliable methods of preventing stroke.

Stroke (cerebrovascular accident) is the third leading cause of death in the United States. An estimated 500,000 Americans are stroke victims each year, and of these 145,000 die. Currently, there are more than 3 million disabled stroke survivors suffering long-term physical and psychological disability.[1] Each year stroke and its after-effects cost America more than 18 billion dollars. Finding cost-effective ways of decreasing stroke morbidity and mortality is therefore of great humanitarian and economic importance. Identification of stroke-prone individuals by simple noninvasive methods during the course of a routine dental evaluation would be a public health measure of great significance.

Atherosclerotic disease (thrombus and embolus formation) in the area where the common carotid artery bifurcates into the internal and external carotid arteries is the most common cause of stroke. On occasion the carotid vessel wall in proximity to the thrombus and/or the thrombus itself may be calcified and visible on routine panoramic dental radiographs.[2] However, plain radiographs cannot distinguish between lesions confined to the vessel wall and those with an intraluminal component. Contrast arteriography has been the "gold standard" for diagnosis for many years but its associated cost, morbidity, and occasional mortality have prohibited its use as a screening modality. Various thermographic systems have historically and sporadically been credited with identifying patients having altered facial skin temperatures because of an intraluminal stenotic plaque.[3-5] However, none of these thermographic studies have used advanced anatomic image analysis programs.

It was the intent of this pilot study to determine the clinical potential of an advanced form of electronic

Received from the School of Dentistry, University of California Los Angeles, Los Angeles, CA.

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0278-2391/94/5212-0007$3.00/0

Clin Oral Invest (2011) 15:563–569
DOI 10.1007/s00784-010-0418-6

ORIGINAL ARTICLE

Prevalence of findings compatible with carotid artery calcifications on dental panoramic radiographs

Stefan Bayer · Ernst-Heinrich Helfgen · Carolin Bös · Dominik Kraus · Norbert Enkling · Sebastian Mues

Received: 22 October 2009 / Accepted: 19 April 2010 / Published online: 30 April 2010

Abstract Cerebrovascular accidents are responsible for killing or disabling more than half a million Americans every year. They are the third leading cause of death in this country. In Germany, the annual stroke incidence reaches 182 cases per 100,000 inhabitants. Stroke there is the fourth leading cause of death. There is a need of finding cost-effective means of decreasing stroke mortality and morbidity. Instruments for early diagnosis are of great humanitarian and economic importance. All possible clinical findings should be taken into account. It is not the demand of this study to present the panoramic radiograph as a screening test method for early diagnosis of atherosclerosis. The aim is to show the potential of this radiograph used in everyday clinical dental practice by the prevalence of radiopaque findings in the carotid region. This study included panoramic dental radiographs of 2,557 patients older than 30 years of age. Fifty-nine percent of the patients were women and 41% were men. The radiographs were adjudged for signs compatible with carotid arterial calcifications appearing as a radiopaque nodular mass adjacent to the cervical vertebrae at or below the intervertebral space C3–4. Of all these radiographs, 4.8% showed radiopaque findings compatible with atherosclerotic lesions. The proportion of women reached 64.8% and that of men reached 35.2%. In accordance to recent literature, the results of this study show that about 5% of the patients show radiological findings compatible with carotid arterial calcifications. Some of these patients at risk for a cerebrovascular accident may be identified in the dentist's office by appropriate review of the panoramic dental radiograph. The suspicion of carotid artery calcifications demands an impetuous referral to an appropriate practitioner who can assist in the control of risk factors and if necessary arrange surgical removal of the carotid arterial plaque. So, the dentist should be aware of this problem and able to make a contribution to stroke prevention.

Keywords Dental panoramic radiograph · Atherosclerosis · Stroke · Stroke prevention · Cardiovascular accident

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Introduction

The account of screening tests and early diagnosis concepts attaches great importance in medical diagnostics. The possibility is offered to take advantage of early intervention and thus of a decrease in mortality and morbidity.

Cardiovascular disease, resulting in stroke or heart attack, is still the leading cause of death of men and women in Germany. Often, they cause an early death below 70 years and thus reduce the life span far below the mean age [1]. In the USA, for example, there were 4.6 million people in 2002 who had to live impaired by the physical and psychological effects of stroke and 12.6 million affected by the effects of heart attack [2].

The arteria carotis communis shows a bifurcation at the carotid bulb. This is where the arteria carotis externa and arteria carotis interna originate. Predilection spots for atherosclerotic lesion are often located at such anatomic structures due to hemodynamic effects of turbulences at the streaming blood, which increase the risk of plaque and intima erosion [3, 4].

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Soft tissue calcifications in the differential diagnosis of opacities superimposed over the mandible by dental panoramic radiography

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Key words: Soft-tissue calcification, rotational panoramic radiography.

Abstract

Not all opacities observed on panoramic dental radiographs are associated with the jaws. Two thousand six hundred and twenty-eight panoramic radiographs obtained from a single dental health clinic were evaluated for the presence of opacities associated with the mandible. Opacities were observed in 4 per cent of cases. The radiographic features of these opacities are presented. The importance of including soft tissue calcifications which may be superimposed over the mandible on panoramic views in the differential diagnosis is discussed. Information on the various soft tissue calcifications which can occur in this region is presented.

(Received for publication April 1989. Revised February 1990. Accepted March 1990.)

Introduction

The ability of panoramic dental radiographs to record the entire maxillo-mandibular region on a single film makes it a valuable diagnostic tool. Ideally, the view will clearly display those objects within the focal trough of the particular machine while at the same time blurring out structures and objects outside the focal trough.[1] Due to the variability of patients it is not possible to construct a machine where only the jaws will be within the focal trough. As a result, radiopaque material present in soft tissue close to the jaws can often be in the zone of sharpness and therefore appear on the panoramic radiograph. These opacities are often apparent when associated with the mandible as there is very little bony superimposition associated with this bone on panoramic views, except perhaps in the midline.[2]

The focal trough of most panoramic units is narrow in the anterior region and broadens posteriorly towards the condyles.[1] Therefore, superimposition is more readily apparent in the posterior region of the mandible.

There are several conditions where calcifications can occur in soft tissues associated with the mandible.[2,3] The tissues involved include both major and minor salivary glands, lymph nodes, blood vessels, muscle and skin. The interpretation of opacities associated with the mandible on panoramic radiographs must, therefore, take into consideration the possibility of soft tissue calcifications superimposed on the bone.

The aim of this study was to determine the prevalence of radiopacities associated with the mandible in panoramic views and to discuss the radiographic

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ARTICLE 3

USE OF PANORAMIC RADIOGRAPHY AMONG AN AMBULATORY DENTAL POPULATION TO DETECT PATIENTS AT RISK OF STROKE

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A B S T R A C T

Panoramic radiographs of 3.6 percent of 1,175 newly accepted dental school patients displayed calcifications in the area of the carotid vasculature. The authors interviewed the patients with calcifications to determine whether they exhibited any recognized atherosclerosis risk factors. One patient had symptoms of atherosclerosis, and a statistically significant correlation was found for obesity among the patients interviewed. The authors concluded that panoramic radiography is useful for identifying some asymptomatic patients with carotid calcifications. These patients should be referred to their physicians promptly for a cerebrovascular work-up as part of an active stroke-prevention strategy.

Cerebrovascular accident, or CVA, also known as stroke, is the third leading cause of death in the United States.[1] Of the approximately 550,000 new stroke cases reported annually in the United States, 150,000 are fatal. The financial cost of the care and rehabilitation of the 3 million people who have survived strokes as of 1994 is estimated at up to $30 billion per year.[2] These numbers illustrate the enormous impact this disease has on public health. Being able to identify cost-effective methods of reducing stroke morbidity and mortality has powerful humanitarian and financial significance. Detecting patients at risk of experiencing CVA early is critical to both the success of preventive strategies and the design of long-term patient management protocols.

Atheromatous plaque in the extracranial carotid vasculature is recognized as the major contributing source of cerebrovascular embolic and occlusive disease.[3] As this vasculature is within the focal trough imaged by dental panoramic radiographs, the panoramic radiographs obtained during routine dental care may be used to identify some patients who are stroke-prone and thus can lead to an intensive investigation of patients' cerebrovascular status and appropriate prophylactic management.

In a previous study, calcifications at the carotid bifurcation were linked to military veterans at risk of experiencing or with a previous history of CVA.[4,5] In 1981, Friedlander and Lande first described the presence of calcifications in the carotid vasculature area on dental panoramic radiographs in approximately 2 percent of 1,000 male veterans attending a dental outpatient clinic.[4] A second study at another Veterans Administration Medical Center, or VAMC, dental outpatient clinic reported that of 182 patients screened, five men and one woman (3.3 percent) displayed calcifications in the carotid artery.[6] Panoramic radiography also detected the presence of calcified carotid artery plaques in 37 percent of 19 white men admitted to a VAMC for treatment of a recent CVA.[7] A survey of an elderly population in a VAMC facility of 134 patients 65 to 88 years of age revealed that 4.5 percent had calcifications of the carotid vasculature.[8]

In each of these studies, most patients exhibited several risk factors for the development of CVA, including increasing age, being

Mineralización de tejidos blandos en radiografías panorámicas

Mineralization of Soft Tissues in Panoramic Radiographies

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Recibido: 5-VI-2019 Aceptado: 9-VII-2019 Publicado Online First: 16-VII-2019

DOI: 10.15517/IJDS.V0I0.38366

RESUMEN

Introducción: La prevalencia de condiciones patológicas que se presentan como radiopacidades a nivel de los tejidos blandos en radiografias panorámicas es una problemática que se da a nivel mundial de la población, siendo este hallazgo radiográfico el diagnóstico inicial de otras afecciones sistémicas. Objetivo: el objetivo de esta investigación fue identificar la frecuencia de radiopacidades mineralizadas que se encuentran a nivel de los tejidos blandos en radiografías panorámicas. Metodología: se analizó 347 radiografías de pacientes mayores de 20 años atendidos en la Clínica Docente Odontológica de la Universidad Católica de Cuenca, Sede Azogues, Ecuador desde diciembre del 2017 hasta mayo del 2018. Resultados: se encontró una prevalencia del 0% de tonsilolitos y anteromas, 1% de ganglios linfáticos calcificados y de antrolitos, 2% de sialolitos, 4% de calcificaciones del ligamento estilohioideo unilateral, 23% de calcificaciones del ligamento estilohioideo bilateral y 65% no presentaron calcificaciones de los tejidos blandos. Conclusión: se pudo identificar que las radiopacidades más frecuentes a nivel de los tejidos blandos es el del proceso estilohioideo calcificado bilateral.

PALABRAS CLAVE

Radiopacidades; Imagen diagnóstica; Tejidos blandos; Calcificaciones; Radiografía; Panorámica.

Acta Odontológica Venezolana - VOLUMEN 49 Nº 3 / 2011
ISSN: 0001-6365 – www.actaodontologica.com

Casos Clínicos:

ATEROMA CALCIFICADO EN CARÓTIDA Y RADIOGRAFÍA PANORÁMICA: REPORTE DE CASO

Recibido para su publicación: 21/04/2010
Aceptado para publicación: 22/07/2010

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Introducción

La ateroesclerosis es una enfermedad caracterizada por la formación de múltiples lesiones focales llamadas placas de ateroma en la pared arterial. Estas placas están compuestas por lípidos y células inflamatorias que pueden calcificarse [1].

El incremento de la presión arterial, los altos niveles séricos de carbohidratos o colesterol así como otros factores como el tabaquismo y la menopausia pueden conducir al desarrollo de daño endotelial y aterogénesis. Específicamente los ateromas de carótida se han asociado con mayor severidad en la enfermedad arterial coronaria, accidentes cerebro-vasculares e infarto cerebral [2]. El accidente cerebro-vascular es causa importante de muerte en varios países, sobre todo en pacientes mayores de 50 años [2].

Cualquier enfermedad que provoque la disminución o la obstrucción de la luz de un vaso como la carótida, disminuirá la cantidad de sangre y oxigeno que llega al órgano irrigado ocasionando una alteración en su funcionamiento. En el caso de la irrigación cerebral, estarían comprometidas importantes funciones que pueden poner en riesgo la vida de un paciente. Por lo tanto, al ser la ateroesclerosis una enfermedad que tarda en dar manifestaciones clínicas (e incluso puede no darlas nunca), se hace necesario la utilización de métodos de diagnóstico precoz que permitan detectar las placas antes de que la irrigación sanguínea se vea comprometida [1,2].

Dentro de los diferentes métodos para diagnosticar ateroesclerosis; la angiografía es considerada el método ideal, sin embargo es un método invasivo que conlleva a varios riesgos derivados de la técnica quirúrgica para llegar a la luz de un vaso [3,4,5]. Otros recursos imagenológicos que incluyen el ultrasonido se han desarrollado para estudiar los grandes vasos como la carótida sin los riesgos de la invasiva angiografía. Así, el Color Doppler (también llamado Duplex scan o Fluxometria laser Doppler) es una prueba diagnóstica cada vez más utilizada para localizar ateromas debido a que es exacta, no-dolorosa, rápida y produce una calidad de imagen similar a la de la angiografía. Y aunque la angiografía puede producir una imagen con mayor detalle, hoy por hoy el Color Doppler es la prueba más indicada por los cardiólogos alrededor del mundo para diagnosticar ateromas ya que no es invasiva [2].

Friedlander y Lande, fueron los primeros en describir la presencia de calcificaciones en el área de la arteria carótida mediante radiografías panorámicas realizadas para el diagnóstico dental de rutina, y encontraron una prevalencia de aproximadamente 2% en una muestra de 1000 pacientes de edad avanzada (6). Numerosos estudios reportan que imágenes radiopacas ubicadas entre la segunda, tercera y cuarta vértebra cervical posteriormente son diagnosticadas como ateromas mediante Color Doppler [1,2,7,8,9,10].

El uso de radiografías panorámicas para identificar ateromas en la carótida es de suma importancia ya que es una radiografía que permite observar fácilmente cualquier calcificación en la zona de las primeras vertebras cervicales, es muy fácil de realizar, no es dolorosa y es mucho más económica que otras

Physiol Rev
84: 1381–1478, 2004; 10.1152/physrev.00047.2003.

Role of Oxidative Modifications in Atherosclerosis

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Stocker, Roland, and John F. Keaney, Jr. Role of Oxidative Modifications in Atherosclerosis. *Physiol Rev* 84: 1381–1478, 2004; 10.1152/physrev.00047.2003.—This review focuses on the role of oxidative processes in atherosclerosis and its resultant cardiovascular events. There is now a consensus that atherosclerosis represents a state of heightened oxidative stress characterized by lipid and protein oxidation in the vascular wall. The oxidative modification hypothesis of atherosclerosis predicts that low-density lipoprotein (LDL) oxidation is an early event in atherosclerosis and that oxidized LDL contributes to atherogenesis. In support of this hypothesis, oxidized LDL can support foam cell formation in vitro, the lipid in human lesions is substantially oxidized, there is evidence for the presence of oxidized LDL in vivo, oxidized LDL has a number of potentially proatherogenic activities, and several structurally unrelated antioxidants inhibit atherosclerosis in animals. An emerging consensus also underscores the importance in vascular disease of oxidative events in addition to LDL oxidation. These include the production of reactive oxygen and nitrogen species by vascular cells, as well as oxidative modifications contributing to important clinical manifestations of coronary artery disease such as endothelial dysfunction and plaque disruption. Despite these abundant data however, fundamental problems remain with implicating oxidative modification as a (requisite) pathophysiologically important cause for atherosclerosis. These include the poor performance of antioxidant strategies in limiting either atherosclerosis or cardiovascular events from atherosclerosis, and observations in

Vol. 117 No. 1 January 2014

Prevalence of calcified carotid artery atheromas in panoramic radiographs of HIV-positive patients undergoing antiretroviral treatment: a retrospective study

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Objective. This study investigated the prevalence of calcified carotid artery atheromas (CCAAs) in panoramic radiographs of HIV-positive patients.
Study Design. A retrospective cross-sectional study was performed to evaluate the presence of CCAA in 300 panoramic radiographs. Qualitative variables were compared using the χ^2 test or Fisher exact test, as needed. The Mann-Whitney or Student *t* test was used for the quantitative variables.
Results. In the studied group, 8.2% presented CCAA. Among these patients, most used lopinavir/ritonavir ($P = .0459$), had a greater mean age ($P = .0081$), and displayed a lower nadir CD4 ($P = .0195$). The use of lopinavir/ritonavir increased the chances of CCAA by approximately 2.8-fold compared with those who did not use medication (odds ratio, 2.79; 95% confidence interval, 1.12-6.95; $P = .045$).
Conclusions. The variables that were associated with the identification of CCAA are compatible with the known atherogenic risk factors in patients with HIV. (Oral Surg Oral Med Oral Pathol Oral Radiol 2014;117:67-74)

Several studies have been published correlating images of carotid atheroma (i.e., detected via panoramic radiographs) with risk factors for the development of cerebrovascular accidents.[1-8] These atheromas often appear near the bifurcation of the carotid arteries, which can be observed in the field of panoramic radiography.[9-11] Previous publications suggest that significant stenosis can exist when atheromatous calcifications are observed in panoramic radiographs.[11,12] The identification of significant stenosis of the carotid artery (luminal narrowing of the vessel above 50%) in asymptomatic patients has significant importance in public health, as shown by the results of several studies demonstrating that the treatment of asymptomatic obstructions decreases the risk of stroke and death. Population screenings to identify asymptomatic lesions are not indicated. However, if identified, these injuries require medical monitoring and treatment.[13,14]

Recognizing the potential benefits of calcified carotid artery atheroma (CCAA) identification in panoramic radiographs (performed routinely for dental treatment), the American Dental Association's Council on Scientific Affairs[15] recommended in 2006 that dentists review the radiographs of their patients for such injuries and that they refer relevant patients for a medical assessment.[15]

The prevalence of carotid calcification identified in panoramic radiographs and documented in the literature varies between 3% and 5% of patients without systemic disease[4,6,11,16-18] and between 20% and 38.8% in populations with known risk factors for atherosclerosis (diabetes, menopause, metabolic abnormalities, cardiovascular disease, chronic renal disease, and hypertension).[2,19-25]

The identification of carotid atheroma in panoramic radiographs also represents an important predictive feature. Individuals with these images have a significantly higher risk of vascular events (myocardial infarction, revascularization procedures, transient ischemic attack, and angina) than do control participants with a similar risk of atheroma. According to Cohen et al.,[26] 2.7 years (mean) after the identification of atheroma, 57%

Statement of Clinical Relevance

The premature occurrence of cardiovascular events in HIV-positive patients and the fact that cardiovascular risk scores often fail to identify young patients at risk lead to the need for additional measures. Panoramic radiographs may be helpful in this context.

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Received for publication May 5, 2013; returned for revision Aug 23, 2013; accepted for publication Aug 28, 2013.

2212-4403/$ - see front matter
http://dx.doi.org/10.1016/j.oooo.2013.08.022

Brief Reviews

Inflammation and the Osteogenic Regulation of Vascular Calcification

A Review and Perspective

Jian-Su Shao, Su-Li Cheng, Justin Sadhu, Dwight A. Towler

Arterial biomineralization processes have been afflicting humans for ≥5 millennia, as realized in 2003 via the computed tomographic imaging of Ötzi, the intriguing "ice mummy" discovered in the Tyrolean Alps.[1] Patchy abdominal atherosclerotic calcification was readily detected in the postmortem of this ≈40-year–old hunter of the early Copper Age, by 2000 years a predecessor of King Tutankhamen.[1] Today, an epidemic of vascular calcification is emerging within our aging and dysmetabolic populace.[2,3] Although vascular calcification was once considered only a passive process of dead and dying cells, work from laboratories worldwide has now highlighted that arterial biomineralization is an actively regulated form of calcified tissue metabolism.[4,5] Moreover, as in skeletal development – where unique biology controls matrix mineralization in membranous bone, endochondral bone, dentin, and enamel,[6,7] mechanistic diversity exists in the pathobiology of vascular calcium deposition.[2,4,5,8] Five common forms of vascular calcification, each possessing unique histoanatomic characteristics and clinical settings with overlapping yet distinct molecular mechanisms, have been described to date[4,5,9] (Table 1). Although we touch on the subject, the reader is referred to other contemporary reviews for in-depth consideration of pathogenic differences.[2,4,5]

In this brief review and perspective, we recount recent data that emphasize inflammation and oxidative stress signaling as key contributors to the pathogenesis of vascular mineral deposition.[10] Furthermore, we highlight differences between the low-density lipoprotein receptor (LDLR)-deficient and apolipoprotein E (apoE)-deficient murine models (Table 2) that help articulate the multifaceted contributions of dyslipidemia, diabetes mellitus, and uremia to arterial calcium deposition.[2,4,11] We end by summarizing the importance of considering these disease stage- and context-specific contributions arterial mineralization when crafting therapeutic strategies to address the disease burden of vascular calcification that increasingly afflicts our patients.[5,12]

Inflammatory Cytokines in the Initiation and Progression of Arterial Calcification: Lessons Learned From LDLR$^{-/-}$ and ApoE$^{-/-}$ Mice

Some degree of vascular inflammation is a frequent concomitant of most forms of arterial calcification.[13,14] Sites of inflammation relevant to disease biology may not only include the atherosclerotic intima and media but also the tunica adventitia.[15–18] Of note, calcification of the elastic lamina with elastinolysis in the absence of overt histological inflammation has been reported,[19–23] and intimal CD68^{+} macrophage accumulation is more commonly associated with atherosclerotic versus medial calcification.[24] However, because calcium phosphate mineral deposition itself elicits inflammatory responses,[25] including tumor necrosis factor (TNF) production by macrophages,[26,27] a primary role for inflammation in the pathogenesis of clinically relevant vascular calcification was unproven until very recently.[28–32] In this section, we review this new data and also highlight distinctions between the LDLR$^{-/-}$ and apoE$^{-/-}$ murine disease models[33] (Table 2) that provide insights into the mechanistic complexities of inflammation-dependent arterial calcium accumulation.

RANKL and Atherosclerotic Calcification

Receptor Activator of Nuclear Factor κB Ligand/ Osteoprotegerin Signaling and Atherosclerotic Calcification

The first robust evidence for the primary contributions of inflammatory cytokine signaling to pathogenesis of vascular calcification arose from the generation and evaluation of the osteoprotegerin (OPG)$^{-/-}$ mouse.[34] OPG-deficient mice develop severe medial and intimal arterial calcification in conjunction with high-turnover osteoporosis driven by excessive osteoclast formation.[34] OPG was first shown to function as an antagonistic "faux receptor" of receptor activator of nuclear factor κB ligand (RANKL), the TNF superfamily member that signals via its receptor activator of nuclear factor κB on monocyte/macrophage progenitors to promote the formation of bone-resorbing osteoclasts.[7,35] In bone, the antagonist OPG is expressed alongside RANKL in the osteoblast lineage, However, OPG is also expressed in vascular smooth muscle cells and endothelial cells of large arteries, a venue where RANKL is normally absent but induced with inflammation.[35] RANKL expression is readily detected in T cells and macrophages near atherosclerotic lesions and within cytokine-stimulated endothelium.[35] Intriguingly, RANKL has been shown recently to promote osteochondrogenic mineral-

Received October 5, 2009; first decision October 20, 2009; revision accepted December 28, 2009.
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(*Hypertension*. 2010;55:579-592.)

Hypertension is available at http://hyper.ahajournals.org DOI: 10.1161/HYPERTENSIONAHA.109.134205

RESEARCH

PANORAMIC RADIOGRAPHY: AN AID IN DETECTING PATIENTS AT RISK OF CEREBROVASCULAR ACCIDENT

ARTHUR H. FRIEDLANDER, D.D.S.; J. DENNIS BAKER, M.D.

A B S T R A C T

Some individuals at risk for a cerebrovascular accident can be identified in the dental office by appropriate review of their panoramic radiographs. Radiographs were evaluated for 304 outpatients, 55 years or older, to check the presence of calcifications associated with artery disease. Three percent of the sample had such opacities.

Cerebrovascular accident (CVA or stroke) is the third leading cause of death in the United States. An estimated 500,000 Americans suffer a CVA each year. Of these, 150,000 die. Currently, there are more than 3 million disabled survivors of CVAs suffering long-term physical and psychological disability.

Each year, CVAs and their after-effects cost the nation more than $25 billion.[1] Identifying stroke-prone individuals during a routine dental evaluation would be a significant public health measure.

Atherosclerotic disease is the most common (85 percent) cause of a CVA. Two-thirds of these are believed caused by thrombus and embolus formation in the region of the carotid bifurcation. We undertook a study to determine the prevalence of calcified carotid atheromata imaged on panoramic radiographs of asymptomatic individuals having an initial dental examination. To confirm the presence and extent of vascular disease, we used color flow Doppler imaging and Doppler spectral analysis for patients with radiographic evidence of carotid arterial disease. We also obtained medical histories and laboratory data to correlate the presence of atherosclerotic disease with risk factors associated with CVAs.

MATERIALS AND METHODS

The study was done at the Veterans Affairs Medical Center in Sepulveda, Calif., a 250-bed, tertiary level care hospital with a complete array of ambulatory clinics. We screened 750 consecutively treated outpatients attending the dental clinic. Imposing inclusion criteria (aged 55 years or older and the ability to sustain a panoramic radiograph) and exclusion criteria (a history of either a transient ischemic attack or a cerebrovascular accident) resulted in a study population of 304 individuals.

A panoramic radiograph was obtained with the patient positioned in the normal manner. A panoramic X-ray system (Panelipse II, Gendex Corp.) was operated at 4 milliamperes with the kilovolt (peak) (range 70-80) dependent on an estimate of the subject's jaw size. Exposed radiographs (Kodak Dental Film/Ektamat: DFG-5 with Kodak X-Omatic Regular intensifying screens, Eastman Kodak) were processed according to the manufacturer's directions. An automatic developer (A/T 2000, Air Techniques, Inc.) was used.

The radiographs were examined in subdued ambient light under optimal viewing conditions using transmitted light from a standard viewing box. They were reviewed by an oral and maxillofacial surgeon (A.H.F.) for the presence of calcifications within the carotid artery. (This clinician reviews more than 2,000 panoramic radiographs each year and has previously reported other data referrable to carotid arterial disease.[2])

Those patients whose radiographs showed calcifications in the region of the carotid vessels underwent color flow Doppler studies (ultrasound)

DENTISTRY AND MEDICINE

Carotid artery atheromas in post-menopausal women

Their prevalence on panoramic radiographs and their relationship to atherogenic risk factors

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ABSTRACT

Background. More than 60 percent of the deaths in the United States attributed to stroke occur in postmenopausal women. As estrogen levels decline, atherosclerotic lesions (that is, atheromas) develop in the region of the carotid bifurcation and have been implicated as the precipitating cause in the majority of these strokes. Atheromas often are calcified and have been detected on the panoramic radiographs of neurologically asymptomatic male veterans; however, similar studies have not been conducted among female veterans.

Methods. The authors assessed panoramic radiographs and medical records of 52 neurologically asymptomatic female veterans (mean age, 70.4 years), with a history of amenorrhea of more than 12 months' duration, for atheromas and risk factors associated with atherosclerosis.

Results. The radiographs of 16 subjects (31 percent) exhibited atheromas located in the neck about 2.0 centimeters inferior and posterior to the angle of the mandible. These findings were confirmed in all instances by the presence of atheromas on anteroposterior cervical spine radiographs. The medical histories of these subjects were heavily laden with atherogenic risk factors (hypertension, 94 percent; body mass index of 27 to 29.9 [characterized as overweight], 25 percent; body mass index of 30 or higher [characterized as obese], 25 percent; smoking more than 15 pack-years, 38 percent; hyperlipidemia, 69 percent; type 2 diabetes mellitus, 21 percent). Hypertension was significantly associated with the presence of atheromas.

Conclusions. Some neurologically asymptomatic women at high risk of developing stroke can be identified in the dental office via panoramic radiography. Women whose X-rays show calcified carotid artery atheromas are almost always hypertensive and have medical histories heavily laden with other atherogenic risk factors.

Clinical Implications. Dentists should refer patients with such calcifications to an appropriate physician for further evaluation and treatment.

JADA CE CONTINUING EDUCATION ARTICLE 4

Thirty-five million women are 55 years of age or older in the U.S. The physiological changes associated with menopause (for example, reduced levels of estrogen) and other processes associated with aging result in women being at a disproportionately high risk of developing stroke, the third leading cause of death in the United States. In 1991, women accounted for almost 61 percent of Americans who died of cerebrovascular disease; however, most epidemiologic studies have centered on identifying prevalence rates and risk factors in men.[1] Stroke and its aftereffects cost the nation more than $30 billion each year.[2] Moreover, identifying stroke-prone older women during the course of a routine dental examination would be a public health measure of great humanitarian significance.

Some women at high risk of developing stroke can be identified in the dental office via panoramic radiography.

The vast majority of strokes in postmenopausal women, as in men, are the result of ischemic cerebral injury caused by atherosclerotic disease (thrombus and embolus formation). Numerous studies have shown that men who have had a stroke almost always have an atherosclerotic lesion at the bifurcation of the common carotid artery. Although far fewer studies have been conducted among women, and almost all have involved small sample sizes, the data implicate high-grade stenotic lesions at the carotid bifurcation as the most likely cause of stroke.[3,4]

Atherosclerotic lesions at the carotid bifurcation

Discrimination between calcified triticeous cartilage and calcified carotid atheroma on panoramic radiography

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The differential diagnosis of calcified atherosclerotic plaque in the extracranial carotid vasculature includes a number of anatomic and pathologic radiopacities. Most of these are readily distinguishable on the basis of location and morphologic features. The calcified triticeous cartilage, however, can be a confounding alternative that is frequently misdiagnosed as a calcified atheroma. This paper describes the radiographic differences between these 2 entities, enabling clinicians to improve their diagnostic acumen when evaluating cervical soft tissue calcifications. (Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2000;90:108-10)

The review of existing panoramic radiographs for the presence of calcified atherosclerotic plaque in the extracranial carotid vasculature has enjoyed considerable interest in recent years.[1-9] During this review process, the clinician must distinguish calcified carotid artery atheromas from anatomic and pathologic radiopacities that lie in close proximity to the vessel. Neighboring anatomic radiopacities include the hyoid bone, epiglottis, and mineralized stylomandibular and stylohyoid ligaments. Pathologic radiopacities that may present in the region of the vessel include calcified thyroid or submandibular gland, sialoliths, phleboliths, calcified lymph nodes, and tonsilloliths. Based on the location and typical morphology of the above-mentioned entities, there is rarely a problem in distinguishing them from calcified atheromas. However, many practitioners are unaware that a calcified triticeous cartilage, or less frequently the superior horn of a calcified thyroid cartilage, may be mistaken for calcified arterial plaque. The purpose of this manuscript is to provide assistance in identifying these variants of normal and to raise clinicians' awareness of the existence of these structures to reduce the likelihood that they will be misdiagnosed as calcified carotid atheromas.

In addition to the thyroid, arytenoid, and cricoid cartilages, 3 smaller pairs of cartilages form part of the upper laryngeal skeleton (Fig 1).[10] The corniculate cartilages are small paired structures situated immediately superior to the arytenoids. Just lateral and superior to the corniculate cartilages, buried in the aryepiglottic folds, are the thin cuneiform cartilages. The round, cord-like lateral

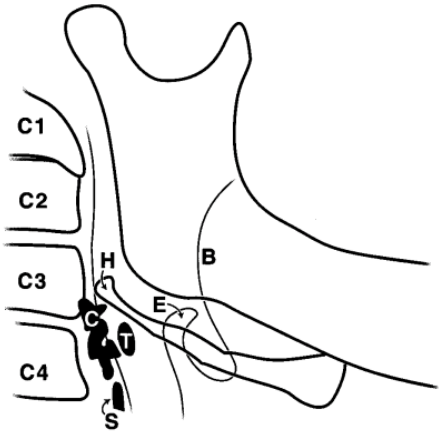

Fig 1. Sketch illustrates morphology and location of calcified triticeous cartilage *(T)*, superior cornu of calcified thyroid cartilage *(S)* and calcified carotid atheroma *(C)* as they appear on panoramic radiograph. Also labeled are the following normal anatomic landmarks: epiglottis *(E)*, greater cornu of hyoid bone *(H)*, soft tissue of base of tongue *(B)*, prevertebral soft tissue *(P)* and parapharyngeal air space *(A)*.

thyrohyoid ligaments form the posterior borders of the thyrohyoid membrane, connecting the posterior aspect of the greater cornua of the hyoid bone with the superior cornua of the thyroid cartilage on each side.[11] The small paired triticeous cartilages are found centrally within the posterior free edge of the lateral thyrohyoid ligaments. These ovoid radiopacities, approximately 2 to 4 mm wide by 7 to 9 mm in length, are usually imaged within the pharyngeal air space adjacent to the superior portion of C4 (Fig 2).[12,13] The word *triticeous* comes from the Latin *triticeus*, meaning resembling a grain of wheat.[14] The function of the triticeous cartilage is unknown,

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Received for publication Oct 18, 1999; accepted for publication Jan 19, 2000.

1079-2104/2000/$12.00 + 0 **7/16/106297**
doi:10.1067/moe.2000.106297

Oral Diseases (2005) 11, 314–317

http://www.blackwellmunksgaard.com

ORIGINAL ARTICLE

Clinicostatistical study of carotid calcification on panoramic radiographs

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OBJECTIVE: The purpose was to evaluate carotid calcifications on panoramic radiographs, and relate to risk factors for vascular diseases.
METHOD: Between 1997 and 2001, 2568 radiographs were retrospectively collected from new patients at Mie University Hospital whose ages ranged from 50 to 70 years. The mean age of the subjects was 62.2 years (men 61.9 years, women 62.3 years). Medical and social data were collected from case notes, and body weight, height, and age of menopause confirmed by telephone interviews.
RESULT: About 106 carotid calcifications were found on the panoramic radiographs of 26 males and 80 females. The ratio of males to females was 1:3.07. The subjects with carotid calcifications had medical histories that included hypertension (27.6%), obesity (21.1%), hyperlipidemia (14.5%), and cardiovascular diseases (13.2%), all with recognized risk factors for atheromas. Of 76 patients who responded to follow up interviews, two (2.63%) died from cardiovascular stroke during an average follow up of 2.43 years.
CONCLUSIONS: The results show carotid calcifications detected on panoramic radiographs can be used to help predict vascular strokes in patients. In cases where calcified carotid artery atheromas are detected, the dentist or oral and maxillofacial surgeon should refer the patient to a specialized physician.
Oral Diseases (2005) 11, 314–317

Keywords: panoramic radiographs; carotid calcification; atheroma

Friedlander and Lande (1981) first reported that calcified carotid artery atheromas (CCAAs) were diagnosable by means of panoramic radiography. Their later study suggested that panoramic radiographs were able to play a significant role in the diagnosis of CCAAs, which could escalate into more serious cerebrovascular disease and heart disease (Cohen *et al*, 2002). These circulatory diseases would manifest themselves an average of 2.7 years after CCAAs symptoms were diagnosed (Cohen *et al*, 2002).

In Japan, there are a few reports on CCAAs detected by panoramic radiographs. However, for Japanese patients with CCAAs identified on panoramic radiographs, the incidence of carotid artery atheromas and the end-points after long-term follow up periods are not clearly established (Fukuta *et al*, 2003; Ohba *et al*, 2003). Therefore, we retrospectively examined panoramic radiographs obtained from new patients of the Department of Oral and Maxillofacial Surgery at Mie University and studied the incidence of CCAAs and the end-point of those patients who had CCAAs. Further, the relationship between CCAAs and gender, life style, and medical history was evaluated.

Materials and methods

Subjects of this study were new patients at the Department of Oral and Maxillofacial Surgery at Mie University who were between 50 and 70 years old (1221 males, 1347 females, or a ratio of 1:1.03 males to females) during the years 1997–2001.

The radiographs were obtained with a X-600/serial number: CPK J105 (Morita, Kyoto, Japan). Panoramic radiography was performed, according to the manufacturer's recommendations (radiographic film; Konika SR-G, kVp; 60–80 V, 5–10 mA, 10–15 s). A radiopaque nodular mass or masses adjacent to the cervical vertebrae at or below the intervertebral space between C3 and C4 were diagnosed as CCAAs. Three dentists interpreted all radiographs and concurred on a diagnosis of carotid artery atheromas. Radiographs that were distorted because of the subjects' movements during the exposure or did not include C3 and C4 were eliminated. Furthermore, the patients who had a history of radiation treatment to the neck and pharyngeal regions were excluded.

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Received 14 May 2004; revised 9 February 2005; accepted 9 February 2005

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ELSEVIER

Vol. 89 No. 4 April 2000

ORAL SURGERY
ORAL MEDICINE
ORAL PATHOLOGY

ORAL MEDICINE

Editor: Jed Jacobson

The prevalence of calcified carotid artery atheromas on the panoramic radiographs of patients with type 2 diabetes mellitus

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Objective. Type 2 diabetes mellitus, which afflicts 15 million Americans, is associated with accelerated cervical carotid artery atherosclerosis and a heightened risk of stroke. This study attempted to determine the prevalence of calcified atherosclerotic lesions in a group of patients with type 2 diabetes mellitus.
Study design. The panoramic radiographs of 49 men (age range, 55 to 81; mean age, 66.2 years) receiving routine dental treatment and insulin for diabetes at a Department of Affairs Veterans clinic were evaluated for calcified atheromas. Age-match controls, free of diabetes, were assessed in a like manner. Statistical comparison of the atheroma prevalence rates was by means of the Fisher exact test, and statistical comparison of atherogenic risk factors was by means of *t* test with Bonferroni adjustment and, where necessary, the Mann-Whitney *U* test.
Results. The radiographs of the diabetics (mean age, 66.9 years) revealed that 20.4% had atheromas whereas those of the controls (mean age, 68.1 years) demonstrated that 4% had atheromas (a statistically significant difference; $P = .0275$). Also statistically significant was the prevalence of atherogenic risk factors (plasma glucose, low density lipoproteins, and serum triglycerides) identified in the diabetic group. The radiographic appearance of the atheromas manifested by both groups of individuals, however, was similar, with the lesions located 1.5-2.5 cm inferior-posterior to angle of the mandible.
Conclusions. People with type 2 diabetes have a greater prevalence of calcified atheromas on their panoramic radiographs than do nondiabetics.
(Oral Surg Oral Med Oral Pathol Oral Radiol Endod 2000;89:420-4)

Type 2 diabetes mellitus (previously known as *non-insulin-dependent diabetes mellitus*) afflicts approximately 15 million Americans.[1] These people are at heightened risk of stroke because hyperglycemia, hyperlipidemia, and a hypertension often associated with the disorder have been implicated as the cause of atherosclerosis of the cervical portion of the carotid artery.[2] Panoramic radiography is capable of discern-ing calcified atherosclerotic lesions (atheromas) in the cervical carotid artery and is frequently used to assess the maxillofacial complex of people with type 2 diabetes because of their known propensity to have oral infections.[3-6] The scientific literature is silent, how-ever, as to the prevalence of atheromas on the radiographs of this high-risk group of patients. Therefore, we undertook a study to determine the prevalence of calcified atherosclerotic lesions among a group of patients with type 2 diabetes.

MATERIAL AND METHODS

Patients studied

The members of the study group were gleaned from 94 consecutively treated outpatients attending the Diabetes Clinic at the Veterans Affairs Los Angeles Ambulatory Care Center. Inclusion criteria for the patients were (1) a diagnosis of type 2 diabetes (ie, in the unmedicated state, having a fasting plasma glucose [FPG] level > 126 mg/dL on 2 separate occasions[7]), (2) daily insulin administration, (3) an age of 55 years or greater, and (4) the ability to sustain a panoramic radiograph. Exclusion criteria for the patients were (1) a history of a transient ischemic attack, (2) a history of a cerebrovascular acci-

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Received for publication June 12, 1998; returned for revision Aug 3, 1998; accepted for publication Apr 19, 1999.

1079-2104/2000/$12.00 + 0 **7/13/99753**
doi:10.1067/moe.2000.99753

The Prevalence of Carotid Artery Calcification on the Panoramic Radiographs in Cappadocia Region Population

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ABSTRACT

Objectives: The aim of this study is to determine retrospectively the presence of carotid artery calcifications (CACs) detected on panoramic radiographs (PRs) in a group of Turkish population. Further, the relationships between CACs and gender, life style, and medical history were evaluated.

Methods: During the years 2004 to 2006, a random sample of 1282 PRs was collected from patients older than 40 years who were being treated by the School of Dentistry, Erciyes University. Of these 1282 PRs, 750 PRs were included in this study. Medical data was collected from the archival records of the dental school.

Results: About 38 (5.06%) CACs were found on the PRs of 12 (4.5%) males and 26 (5.4%) females. The CAC prevalence was not significantly different between the males and females (P=0.583). These calcifications were unilateral in 26 (68.4%) and bilateral in 12 (31.6%) subjects. Of those in the positive group, there were 12 subjects (31.58%) with hyperlipidemia, 12 subjects (31.58%) with hypertension, 7 subjects (18.4%) with diabetes mellitus, 6 subjects (15.8%) with cardiovascular disease, and 6 subjects (15.8%) with smoking history.

Conclusions: This study has the highest CACs prevalence in comparison to the other studies. Therefore, dentists caring for subjects with dental problems should carefully evaluate their PRs for the evidence of CACs, and refer them for medical evaluation as indicated. So, incidental findings could provide life-saving information. (Eur J Dent 2007;1:132-138)

Key words: Panoramic radiograph; Carotid artery calcification; Atherosclerosis, Risk factors

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Health, 2014, 6, 1933-1940
Published Online August 2014 in SciRes. http://www.scirp.org/journal/health
http://dx.doi.org/10.4236/health.2014.615226

Scientific Research

Evaluation of Positive Predictive Value for Digital Panoramic Radiography in Comparison to Ultrasound in the Diagnosis of Calcified Carotid Atheroma

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Received 4 June 2014; revised 20 July 2014; accepted 2 August 2014

Abstract

Aim: Detection of calcified carotid atheroma (CCA) has an important role in reducing the incidence of Cerebro Vascular Accident (CVA). The aim of this study was to evaluate efficacy of panoramic digital radiography in detecting atherosclerosis. Methods: It is descriptive-analytical diagnostic study. The people (22 to 62 years old) were referred to a radiology clinic to perform panoramic radiography for diagnosis of CCA. Individuals who were suspected were introduced to the radiology department of dental school to undergo ultrasound evaluation to CCA. For the 41 patients (55 sides), ultrasound was performed. For data analysis, the Chi-square and Fisher's exact test were used. Results: The prevalence of CCA was 2.43%. The PPV of digital panoramic was 45.5%. There was no significant relationship between age (P = 0.14) and sex (P = 0.539) and PPV of digital panoramic. The PPV of digital panoramic was significantly associated with hypertension (P = 0.032). Conclusion: It seems that panoramic can be used to screen patients with a history of hypertension for atherosclerosis.

Keywords

Radiography, Panoramic, Digital, Ultrasonography, Atherosclerosis

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How to cite this paper: Ezoddini-Ardakani, F., Mirzaei, M., Nayer, S., Besharati, S. and Moeini, M. (2014) Evaluation of Positive Predictive Value for Digital Panoramic Radiography in Comparison to Ultrasound in the Diagnosis of Calcified Carotid Atheroma. *Health*, **6**, 1933-1940. http://dx.doi.org/10.4236/health.2014.615226

ACTA
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CROATICA
www.ascro.hr

Acta stomatol Croat. 2016;50(3):230-234.
DOI: 10.15644/asc50/3/5

IZVORNI ZNANSTVENI RAD
ORIGINAL SCIENTIFIC PAPER

Jéssica Rodrigues da Silva Noll Gonçalves[1], Juliana Laís Yoshie Yamada[1], Cristina Berrocal[1], Fernando Henrique Westphalen[1], Ademir Franco[1], Ângela Fernandes[1]

Prevalencija patoloških nalaza na panoramskim radiogramima: Kalcificirani aterom karotidne arterije

Prevalence of Pathologic Findings in Panoramic Radiographs: Calcified Carotid Artery Atheroma

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Sažetak

Svrha rada: Autori su željeli procijeniti prevalenciju slika koje mogu upućivati na kalcificirane ateroma karotidne arterije (ISCCAA) na panoramskim radiogramima stomatoloških pacijenata. **Materijali i metode:** Uzorak se sastojao od 8338 panoramskih radiograma pacijentica (n = 5 049) i pacijenata (n = 3 289) u dobi od 4 do 94 godine. Panoramski radiogrami analizirani su zbog ISCCAA-e. Dobiveni podatci statistički su povezani sa spolom i dobi. **Rezultati:** ISCCAA je pronađena na 579 radiograma (6,9 %). Nije bilo statistički značajne razlike između muškoga i ženskoga spola (p > 0,05). ISCCAA je bila prevalentnija kod pacijenata u srednjoj dobi, dakle od 50 godina (p < 0,05). **Zaključak:** Prema dobivenim nalazima može se zaključiti koliko je, u sklopu kliničke dentalne prakse, važna rana dijagnoza potencijalnih slučajeva ISCCAA-e s pomoću panoramskih radiograma.

Zaprimljen: 9. travnja 2016.
Prihvaćen: 28. lipnja 2016.

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Ključne riječi
karotidne arterije; arterioskleroza; plak; kalcifikacija, vaskularna; panoramska rendgenska snimka;

Uvod

Ateroskleroza je upalna kronična bolest koju obilježava zadebljanje, sužavanje i/ili gubitak elastičnosti stijenke arterija (1, 2). Njezino multietiološko podrijetlo uključuje biokemijski fenomen koji uzrokuje patološku kalcifikaciju arterija (3). Te kalcifikacije zovu se ateromi i potječu od lokalne proliferacije fibroblasta te odlaganja kalcija, a najčešće se nalaze na mjestima gdje se račvaju karotidne arterije (4) i gdje je brzina protoka krvi smanjena pa se povećava izlaganje aterogenim česticama sklonima kalcifikaciji (5).

Klinička važnost ateroma očituje se u njihovu otkidanju od arterijskih stijenki i o uključivanju u krvotok te u potpunom začepljivanju arterije na užim dijelovima, što može rezultirati cerebrovaskularnim inzultom (2, 6 – 8). Gotovo 20 posto ishemijskih cerebrovaskularnih inzulta uzrokovano je otkidanjem ateroma iz karotidne arterije (9).

Doktori dentalne medicine imaju iznimno važnu zadaću u sprječavanju cerebrovaskularnih inzulta jer mogu, zahvaljujući panoramskim radiogramima, rano uočiti i dijagnosticirati kalcificirani arterijski aterom (ISCCAA). Ti radiogrami uo-

Introduction

Atherosclerosis is an inflammatory chronic disease characterized by the thickening, narrowing and/or or loss of elasticity of artery walls (1,2). The multi-etiological origin of atherosclerosis involves a biochemical phenomenon that creates pathological arterial calcifications (3). These calcifications, now called atheromas, originate from the local proliferation of fibroblasts and deposition of calcium. Atheromas are particularly common in the bifurcation of the carotid artery (4), in which the blood flow velocity is reduced increasing the exposure to atherogenic particles that are prone to calcification (5). The clinical importance of atheromas mainly consists in their potential disruption from artery walls following the blood flow, completely obstructing the artery in more narrowed regions, and consequently leading to cerebrovascular accidents (2,6-8). Nearly 20% of the ischemic cerebrovascular accidents are caused by disrupted atheromas in the carotid artery (9).

Dentists play an important role by preventing cerebrovascular accidents through the early diagnosis of images sug-

JOURNAL OF APPLIED ORAL SCIENCE

J Appl Oral Sci. *2009;17(1):45-8*

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DIAGNOSTIC AGREEMENT BETWEEN PANORAMIC RADIOGRAPHS AND COLOR DOPPLER IMAGES OF CAROTID ATHEROMA

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Received: September 26, 2007 - Modification: March 13, 2008 - Accepted: August 13, 2008

ABSTRACT

The aim of this study was to investigate the agreement between diagnoses of calcified atheroma seen on panoramic radiographs and color Doppler images. Our interest stems from the fact that panoramic images can show the presence of atheroma regardless of the level of obstruction detected by color Doppler images. Panoramic and color Doppler images of 16 patients obtained from the archives of the Health Department of the city of Valença, RJ, Brazil, were analyzed in this study. Both sides of each patient were observed on the images, with a total of 32 analyzed cervical regions. The level of agreement between diagnoses was analyzed using the Kappa statistics. There was a high level of agreement, with a Kappa value of 0.78. In conclusion, panoramic radiographs can help detecting calcifications in the cervical region of patients susceptible to vascular diseases predisposing to myocardial infarction and cerebrovascular accidents. If properly trained and informed, dentists can refer their patients to a physician for a cardiovascular evaluation in order to receive proper and timely medical treatment.

Key words: Radiography, panoramic. Carotid artery, external. Carotid artery diseases. Calcinosis.

INTRODUCTION

Increased blood pressure, high serum levels of cholesterol or carbohydrates, and other factors may lead to endothelial damage and consequent atherogenesis[6]. The presence of carotid atheroma is associated with the severity of coronary artery disease (CAD) and cerebrovascular accident (CVA) or cerebral infarction.

Among the different methods to diagnose atherosclerotic diseases, angiography is considered the gold standard. However, because it is an invasive method, complications may occur[5,24,25]. Therefore, color Doppler imaging, also called laser Doppler fluxometry or duplex scan, has been increasingly used to diagnose atheroma because it is a fast, accurate and painless method of diagnosis. Color Doppler can be considered as a gold standard because the results obtained with this imagining method are similar to those obtained with angiography, with the advantage that color Doppler is a noninvasive method[7,15,16,19,23].

Panoramic radiographs, used as a complementary examination resource by dentists, can often show the presence of carotid atheroma[2,22]. Most studies found in dental literature report that calcifications between the 2nd, 3rd and 4th vertebrae first seen on panoramic radiographs are further confirmed as atheroma by color Doppler[3,8,10,20].

Studies have investigated the possibility of identifying carotid artery calcifications on panoramic radiographs, on many occasions especially in asymptomatic cases. Therefore, patients can be referred to a cardiologist or a neurologist for further investigation[9,12].

The use of panoramic images to assess carotid atheroma is important because they often show the presence of calcifications. Panoramic radiographs are easy to take and have a low cost compared to other imaging methods. In addition, a complete radiographic examination of the both arches can be carried out with only one x-ray exposure and a relatively lower radiation dose[2,21].

Our interest stems from the fact that panoramic radiographs can show the presence of carotid calcification regardless of the degree of obstruction. Therefore, the purpose

Dentomaxillofacial Radiology (2006) 35, 266–270

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RESEARCH

Digital panoramic radiography: a reliable method to diagnose carotid artery atheromas?

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Objectives: The aim of the present study is to evaluate the panoramic radiographs of 4106 patients for carotid artery atheromas (CAAs) and to correlate our findings with the literature.
Materials and methods: The digital panoramic radiographs of 4106 dental patients (2428 female, 1678 male) were evaluated. Radiographs of patients 40 years of age or older were randomly chosen from a computer database. CAA findings were defined as radiopaque masses adjacent to the cervical vertebrae at or below the intervertebral space between C3 and C4 on the panoramic radiograph. The patients who had CAA findings were contacted by telephone, and some of them agreed to further evaluation. Evaluation included carotid ultrasound, ECG, echocardiography and treadmill exercise testing at the Department of Cardiology.
Results: Of 4106 patients, 88 patients (2.1%; 70 female, 18 male) had one or more radiopaque mass detected on digital images. All 88 patients with CAA findings were contacted by telephone and 23 agreed to further evaluation at the university hospital. Of these 23 patients, 8 (34.7%) had CAAs on carotid ultrasound and 15 (65.3%) had normal carotid arteries. From these eight patients with CAAs on Doppler ultrasound, 7 (30.4%) had plaques that were not haemodynamically significant and only one (4.3%) had significant plaque. The patient with severe carotid artery stenosis consequently underwent endarterectomy operation. There were no statistically significant differences between male and female in CAAs ($P > 0.05$).
Conclusion: Digital panoramic images may have some diagnostic value for detecting CAAs and this early diagnosis could potentially increase the length and quality of life for people with CAAs.
Dentomaxillofacial Radiology (2006) **35,** 266–270. doi: 10.1259/dmfr/50195822

Keywords: radiography, digital, dental; stenosis; carotid artery

Introduction

Atheromatous plaque accumulates on the posterior wall of the internal carotid artery and extends downwards into the bifurcation of the common carotid artery. Diabetes mellitus, hypertension and exposure to cigarette smoke are all known to cause functional and morphological injury to the endothelium and thus initiate the development of plaque. Atherosclerotic plaque consists of focal fat, cholesterol in the intima layer of arteries and calcium salt deposits. This build-up roughens the arterial endothelium, creating a thickened irregular surface that can cause turbulent blood flow around the build-up. This surface can also trigger the development of clots that narrow the arterial lumen. In this situation, the body releases enzymes to regulate blood flow, which causes an embolus to break free from the plaque surface.[1,2]

One half of all strokes are believed to be the result of atherosclerotic disease and are one of the main causes of death.[3] Millions of people suffer strokes each year. Atherosclerosis is a progressive process and early diagnosis is crucial. Unfortunately, the first clinical manifestation of carotid artery atheroma (CAA) is often a completed stroke that occurs when treatment is too late. However, CAAs can be detected early. Panoramic radiographs, obtained during professional dental examinations, are a potential method for early detection. Their discovery can provide life-saving information. Whenever CAAs are detected on a panoramic radiograph, further diagnostic examination should be recommended. Early detection of CAAs is also economically significant. The cost of treatment for atherosclerosis is lower than for treatment

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Received 25 July 2005; revised 1 November 2005; accepted 22 November 2005

Journal of
Dental Research, Dental Clinics, Dental Prospects

Original Article

Doppler Sonography Confirmation in Patients Showing Calcified Carotid Artery Atheroma in Panoramic Radiography and Evaluation of Related Risk Factors

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Received: 26 Jun 2011; Accepted: 18 October 2011
J Dent Res Dent Clin Dent Prospect 2012; 6(1):6-11 | doi: 10.5681/joddd.2012.002
This article is available from: http://dentistry.tbzmed.ac.ir/joddd

Abstract

Background and aims. The purpose of this study was to identify patients at the risk of cerebrovascular attack (CVA) by detecting calcified carotid artery atheroma (CCAA) in panoramic radiography and evaluating their risk factors.

Materials and methods. A total of 960 panoramic radiographs of patients above 40 years old were evaluated. Doppler Sonography (DS) was performed for patients who showed calcified carotid artery atheroma (CCAA) in panoramic radiography in order to determine the presence of CCAA and the degree of stenosis. Cardiovascular risk factors in both groups of patients with CCAA (12 subjects) and without CCAA (3 subjects) were compared using a questionnaire filled out by the patients. Statistical analysis including Fisher and independent t-test applied for data analysis.

Results. Fifteen patients (30 sides) showed calcification in their panoramic radiographs, and underwent DS which revealed CCAA in 16 sides (12 patients). Two patients (13.33%) showed stenosis greater than 70%. Among the risk factors, only age showed a significant association with the occurrence of carotid calcified atheroma (P=0.026).

Conclusion. Considering the results, dentists should refer especially elderly patients with radiographically identified atheromas for further examinations, as asymptomatic CCAA might be associated with high degrees of stenosis.

Key words: **Calcified carotid artery atheroma, Doppler sonography, panoramic radiography.**

Introduction

Carotid atheroma is an atherosclerotic process occurring along the lumen walls of the common carotid artery near its bifurcation. Pieces of the atheroma may ulcerate and become detached to form an embolus that can occlude smaller intracerebral arteries, causing stroke.[1] Approximately 80% of the

Brief Report

Successful Carotid Endarterectomy For Cerebrovascular Insufficiency

Nineteen-Year Follow-up

Michael E. DeBakey, MD

• This is believed to be the first successful case of thromboendarterectomy for cerebrovascular insufficiency caused by atherosclerotic occlusion of the carotid artery, as well as the longest follow-up study. At the time of the patient's death from coronary occlusion, 19 years after operation, he had no cerebrovascular symptoms, and there was clinical evidence of maintenance of the restored circulation in the carotid artery.

(*JAMA* 233:1083-1085, 1975)

IN 1953, I saw a patient with symptoms of circulatory insufficiency in the left carotid arterial bed and left side of the brain, which, on the basis of the clinical manifestations, I believed to be due to arteriosclerotic occlusion of the left common carotid artery. Because of the good results that I had had with both endarterectomy and graft replacement for segmental atherosclerotic lesions producing circulatory insufficiency in other arteries, particularly those in the legs, I believed that the same procedure would restore normal circulation in the left carotid artery in this patient. The operation was successfully performed, and as far as I have been able to determine, represents the first successful case of thromboendarterectomy of the carotid artery. The patient was observed periodically until his death from coronary occlusion 19 years after carotid endarterectomy. Throughout that time, the restored circulation in the carotid arteries was maintained. For these reasons, the case is reported in detail.

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Report of a Case

A 53-year-old male bus driver was first seen July 29, 1953, because of intermittent episodes of weakness of the right arm and leg, hesitancy and difficulty in speaking, and difficulty in writing clearly. About 2½ years before, he had experienced for the first time weakness and numbness of the right arm associated with expressive aphasia. Thereafter, he had had a number of these episodes, the most recent several weeks before admission. He had also noted some decrease in visual acuity.

At the time of admission to the hospital one week after his initial visit, the patient was well oriented but somewhat sluggish, with mild hesitancy in speech and slight slurring. The blood pressure was 140 mm Hg systolic and 90 mm Hg diastolic. Pulsations in both wrists were normal. Pulsation in the carotid artery on the right was normal, but on the left side it was extremely weak. Similarly, the superficial temporal pulse on the right was easily palpable, whereas that on the left was only questionably palpable. The tendon reflexes on the right were hyperative. Hoffmann, Babinski, and Chaddock signs were positive on the right. No evidence of cerebellar disturbance was observed on examination of the limbs. Sensation was normal on both sides. The right femoral, popliteal, and pedal pulses were absent. On the left side, the femoral pulse was palpable but diminished, and the pedal pulses were weakly palpable.

No abnormality was detected in the electrocardiogram. Roentgenograms of the chest and abdomen showed no significant abnormalities. The electroencephalogram provided minimal evidence of focal abnormality in the left temporal region.

A diagnosis was made of occlusion of the left internal carotid artery originating at the bifurcation of the common carotid and producing cerebrovascular insufficiency. Since published reports had indicated that such lesions may be well localized at the bifurcation of the common carotid artery, it was believed that normal circulation could be restored in the distal internal carotid artery by thromboendarterectomy or resection of the diseased segment and replacement with a graft, provided that the distal segment of the internal carotid artery was patent.

The projected surgical treatment was discussed with the patient, who was told that, as far as I knew, this operation had never been successfully performed on the carotid artery, but that I had had considerable favorable experience with both endarterectomy and graft replacement in other arteries, especially for circulatory insufficiency of the legs. He was also told

The New England
Journal of Medicine

Volume 325 AUGUST 15, 1991 Number 7

BENEFICIAL EFFECT OF CAROTID ENDARTERECTOMY IN SYMPTOMATIC PATIENTS WITH HIGH-GRADE CAROTID STENOSIS

NORTH AMERICAN SYMPTOMATIC CAROTID ENDARTERECTOMY TRIAL COLLABORATORS*

Abstract *Background.* Without strong evidence of benefit, the use of carotid endarterectomy for prophylaxis against stroke rose dramatically until the mid-1980s, then declined. Our investigation sought to determine whether carotid endarterectomy reduces the risk of stroke among patients with a recent adverse cerebrovascular event and ipsilateral carotid stenosis.

Methods. We conducted a randomized trial at 50 clinical centers throughout the United States and Canada, in patients in two predetermined strata based on the severity of carotid stenosis — 30 to 69 percent and 70 to 99 percent. We report here the results in the 659 patients in the latter stratum, who had had a hemispheric or retinal transient ischemic attack or a nondisabling stroke within the 120 days before entry and had stenosis of 70 to 99 percent in the symptomatic carotid artery. All patients received optimal medical care, including antiplatelet therapy. Those assigned to surgical treatment underwent carotid endarterectomy performed by neurosurgeons or vascular surgeons. All patients were examined by neurologists 1, 3, 6, 9, and 12 months after entry and then every 4 months. End points were assessed by blinded, independent case review. No patient was lost to follow-up.

Results. Life-table estimates of the cumulative risk of any ipsilateral stroke at two years were 26 percent in the 331 medical patients and 9 percent in the 328 surgical patients — an absolute risk reduction (±SE) of 17±3.5 percent ($P<0.001$). For a major or fatal ipsilateral stroke, the corresponding estimates were 13.1 percent and 2.5 percent — an absolute risk reduction of 10.6±2.6 percent ($P<0.001$). Carotid endarterectomy was still found to be beneficial when all strokes and deaths were included in the analysis ($P<0.001$).

Conclusions. Carotid endarterectomy is highly beneficial to patients with recent hemispheric and retinal transient ischemic attacks or nondisabling strokes and ipsilateral high-grade stenosis (70 to 99 percent) of the internal carotid artery. (N Engl J Med 1991; 325:445-53.)

CAROTID endarterectomy was introduced in 1954 as a logical procedure for the prevention of ischemic stroke distal to carotid-artery stenosis.[1] Although the first randomized trials of its effectiveness had negative results,[2-4] surgeons continued to perform carotid endarterectomy and began to report lower rates of perioperative complications.[5,6]

The number of patients undergoing endarterectomy in hospitals in the United States (other than Veterans Affairs hospitals) rose from 15,000 in 1971 to 107,000 in 1985.[7] However, continuing uncertainty about the efficacy of the operation was reflected in marked geographic variation in the rates of endarterectomy.[8] Adding to this uncertainty was the decline in the number of first and fatal strokes,[9-11] the influence of risk-factor management in reducing strokes,[12-14] and emerging recognition of the efficacy of antiplatelet drugs in preventing stroke.[15] When a randomized trial demonstrated that extracranial–intracranial bypass was ineffective in preventing stroke,[16] this presented an opportunity to reexamine the current efficacy of carotid endarterectomy as performed in North America, and several randomized trials were begun in both symptomatic and asymptomatic patients,[17] complementing the European Carotid Surgery Trial already under way.[18] This report describes the first definitive results of this new round of trials of carotid endarterectomy.

METHODS

A full description of the methods of the study has been published elsewhere.[19] The key features of the conduct of the trial were as follows.

Center Eligibility

The study was conducted at 50 centers in the United States and Canada. Each center had a rate of less than 6 percent for stroke and death occurring within 30 days of operation for at least 50 consecutive carotid endarterectomies performed within the previous 24 months, and each had obtained approval of the research protocol from its local institutional review board.

Patient Eligibility

To be eligible for the trial, patients had to give informed consent, be less than 80 years old, and have had a hemispheric transient ischemic attack (distinct focal neurologic dysfunction) or monocular

*The collaborators in this trial are listed in the Appendix.

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Supported by a grant (R01-NS-24456) from the National Institute of Neurological Disorders and Stroke.

The New England Journal of Medicine

Owned and Published by the
Massachusetts Medical Society

CAROTID ENDARTERECTOMY FOR ASYMPTOMATIC CAROTID STENOSIS

ISCHEMIC stroke remains one of the most common devastating illnesses in developed countries, ranking third as the cause of death and extracting a huge socioeconomic toll because of the permanence of the disability experienced by many of those who survive. Possible medical and surgical measures to prevent stroke therefore assume paramount importance. Their potential impact demands that their value be affirmed or denied with all the certainty that can be brought to bear by modern methods.

Carotid endarterectomy has been proposed as a stroke-preventing measure and employed with increasing enthusiasm since its introduction in 1954. The frequency of its application in non–Veterans Affairs hospitals in the United States peaked in 1985 at 107,000 patients.[1] Thereafter, growing uncertainty about indications for its use brought a decline, so that in 1990 only 68,000 operations were performed (Dyken ML, Pokras R: personal communication). Within a decade of its introduction, the technique of the randomized clinical trial was used to evaluate this operation. Two small trials failed to establish the benefit of surgery, both because of early flaws in the discipline of the randomized, multicenter trial and because the rates of perioperative complications defeated any attempt to demonstrate the superiority of surgical care.[2,3]

In 1991 the *Journal* published a report from the North American Symptomatic Carotid Endarterectomy Trial (NASCET) that clearly showed that under specified conditions, surgery in addition to medical care was superior to medical care alone.[4] This conclusion applied only to patients with carotid stenosis proved by arteriography to equal or exceed 70 percent when the narrowest diameter of the stenosed lesion was compared with the normal artery beyond the carotid bulb.[4] Study patients had to have had recent symptoms of transient ischemic attacks or nondisabling stroke in the territory of the narrowed artery. In the same year, a similar European trial in symptomat-

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