Research Article

Osteoporosis and Cardiovascular Disease are Linked to Sharp Reduction in Mitochondrial Synthase, Phospholipase, COX2 Enzymes, and TXA2 Alpha Subunits Productions, and Linked to the Feedback of VEGF-A Alpha Subunits from TXA2 Productions

Ashraf Marzouk El Tantawi*

***Corresponding Author: Dr. Ashraf Marzouk El Tantawi** Biomedical molecular studies. Cancer and viral infections. Canada, Toronto, Goldwin Av. Egypt, Giza, Abd elmonem R.

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Abstract

Osteoarthritis is a sharp reduction in Thymine nucleotides that reflect sharp reductions in mitochondrial synthase, phospholipase, and COX2 enzymes lead to a reduction in TXA2 alpha subunits productions which will lead to a reduction in feedback for VEGF-A alpha subunits productions, lead to decreasing in PPARs reactivities, and increasing in catabolic processes by the effects of VEGF-B on joints, and on muscles.

Osteoprotegerin (OPG), which is a soluble glycoprotein, and is distributed in tissues including bone-building cells (osteoblasts) are regulating both bone and vascular anabolism but under the control of the anti-inflammatory engine and its tools productions which are TXA2 alpha subunits and then it's feedback VEGF-A alpha subunits and cells. Where Osteoprotegerin is the alpha subunit that is activated by TXA2 alpha subunits productions.

A sharp drop in the productions of TXA2 alpha subunits due to a sharp reduction of the activities of phospholipase, and synthase and COX2 enzymes on inflammations molecules, will lead to a sharp reduction in VEGF-A alpha subunits productions which occurred due to the feedback from the TXA2 alpha subunits which produced from the effect of the active mitochondrial enzymes on toxicities molecules and inflammations molecules.

Where the so imp feedback from TXA2 alpha subunits to stimulate the production of VEGF-A alpha subunits is considered to be an essential imp step for the productions of VEGF-A alpha subunits and re-strengthen anti-inflammatory engine and its micro alpha subunits tools, for re back reproduce endothelin-1 and reactivate G-actin filaments. Where TXA2 & VEGF-A alpha subunits are having so strong basic roots with chromosome composition functions, where, if histone increased in nucleus gene compositions, will cause delays in many of the productive metabolic cycles in living cells which are controlled originally from the basics of the chromosome compositions.

The mitochondrial synthase enzymes are involved in pyrimidine synthesis specifically from purine nucleotides, which are considered to be the rate of pyrimidine limiting biosynthesis in tissue cells.

Sharp Reductions in anti-inflammatory TXA2 alpha subunits productions which have the so imp feedback steps to produce VEGF-A subunits, is the main reasons for the reduction in VEGF-A alpha subunits productions and inhibition in its reverse cycles for stimulating G-actin filaments and ET-1 reactivities, which are the main reason for osteoporosis with the cardiovascular pathogenic disease.

Where sharp drops in TXA2 and VEGF-A alpha subunits productions will lead to accumulations of toxicities and impurities in interstitium fluid and blood and will lead to re-feed back from toxicities molecules to reform unknown organic and inorganic toxic molecules that lead precipitations in arteries and forming unknown inorganic molecules in interstitium fluid that will appear to us are produced from bone cells and Cartilage.

Osteoarthritis is a sharp reduction in Thymine pyrimidine nucleotides synthesis from purine nucleotides which in necessary for Leu synthesis. Pyrimidine and Purine nucleotide metabolic synthesis and their limit of regulations have strong relations to osteoporosis pathogens.

Purpose of this Work

Osteoporosis and cardiovascular disease are linked to a sharp reduction in mitochondrial synthase, phospholipase, COX2 enzymes, and TXA2 alpha subunits productions.

Then mitochondrial enzymes: TXA2 synthase, COX2, and phospholipase enzymes are so necessary for acting on inflammations molecules which found in interstitium fluid and blood for TXA2 subunits productions, and then for feedback for VEGF-A subunits synthesis, where the last VEGF-A subunits are so necessary for PPARs genes reactivations, and for re-stimulation the endothelin-1 synthesis and re stimulations the G_actin filaments for completing the anti-inflammation cycles and processes.

Introduction

Osteoarthritis (OA) is the most common chronic disease of human joints specifically in the case of the weakness in immune efficiency. OA is the basis of the catabolic cycles of the pathologic cases changes involves in all the tissues including sharp reductions in anti-inflammatory alpha subunits which necessary for re-stimulating the joints recoveries; that, at an early stage, it has the nature of inflammations with varying degrees of severity, that in the reductions of anti-inflammatory main short cycles and tools will lead to change anabolic cycles in all tissues including joints to mostly catabolic processes and cycles, and its pathogenic changes can grow and increases, exactly as cancer if it's main roots, will not be recovered through re-strengthen full anti-inflammatory cycles.

Muscle weakness and its relaxations have strong links with the basis of osteoporosis pathogen, and with the weakness and relaxation of arteries compositions and activities, and the dropping in blood pressure, where the first definition is declared as a sharp decline in the functions of the tissue cells mitochondria and so sharp decline in the productions of mitochondrial enzymes from its active membrane.

Materials

Osteoblasts
TXA2 alpha subunits
VEGF-A alpha subunits
VEGF-B subunits

Mitochondrial enzymes: 1. TXA2 synthase, 2. phospholipase enzyme, 3. COX2,
Osteoprotegerin (OPG).
AMPK, TOR protein
Endothelin_1
G_actin filaments isoforms
TNFα TGF-β
Osteogenic cells
G protein alpha 13
(Thymine) pyrimidine and purines
Collagen bone matrix
Amidotransferase (PPAT) enzymes

Methods and Results

The increasing of histone and glutamine in blood and tissues interstitium will reflect specifically the reductions of leucine amino acids synthesis and activities, which are so necessary for mostly all metabolic reactivities in tissues cells, and joints through the re-synthesis of effective sestrin Leu active carrier tools which are having its own active AMPK and TOR protein and are energized by ribosomal ATPase and G_actin ATPase proper activities.

Osteogenic cells due to reductions or deficiency in Leu (CTT) amino acids availabilities is producing gamma-Glutamine peptides that in normal cases: gamma glutamate (GAA) protein is supposed to give feedback to produce alpha-Leu (CTT) protein through translations processes and that will be done by mitochondrial synthase enzymes and phospholipase enzymes where have been synthesized from the mitochondrial membrane, where synthetase enzymes responsible for reforming and producing thymine nucleotides from purines, which by the mechanism of synthetase enzymes **(**6**)**.

In the case of Osteoporosis, the Collagen bone matrix that has been expressed by calcified plaque is a protein containing gamma-carboxy-glutamate, which is formed due to full reductions in carboxyl-phosphate-synthetase enzymes, which has two forms in mitochondria and cytoplasm **(**6**)**.

Due to the absence of alpha-subunits, the beta (in trace percentage compared to gamma peptides subunits & gamma subunits peptides will be always produced due to catabolic processes and will always following catabolic processes, and will always miss anabolic processes which should be done through feedback from beta and gamma subunits peptides to produce alpha subunits.

Alpha cells or subunits are the main for Bone synthesis, and the feedback from beta and Gamma peptides subunits to produce alpha peptides subunits are so necessary steps to be done by the effect of mitochondrial enzymes including phospholipase and synthase enzymes. The synthase mitochondrial enzymes are involved in pyrimidine synthesis specifically from purine nucleotides, which are considered to be the rate of pyrimidine limiting biosynthesis in tissue cells.

Sharp Reductions in anti-inflammatory TXA2 alpha subunits productions which have the so imp feedback steps to produce VEGF-A subunits, is the main reasons for the reduction in VEGF-A alpha subunits productions and inhibition in its reverse cycles for stimulating G-actin filaments and ET-1 reactivities, which are the main reason for osteoporosis with the cardiovascular pathogenic disease.

Where sharp drops in TXA2 and VEGF-A alpha subunits productions will lead to accumulations of toxicities and impurities in interstitium fluid and blood and will lead to re-feed back from toxicities molecules to reform unknown organic and inorganic toxic molecules that lead precipitations in arteries and forming unknown inorganic molecules in interstitium fluid that will appear to us are produced from bone cells and cartilages.

The full reductions in leucine amino acids activities will reflect the full reductions in Thymine nucleotides, which need mitochondrial enzymes to be in proper productive conditions to act on toxic & impurities molecules and on inflammations products to produce TXA2 alpha subunits which are the main signals rings for feedback for VEGF-A alpha subunits productions, which are the main control root for resynthesis the VEGF-B, and the stimulations of PPARs proliferation generative genes which will be done by VEGF-A alpha subunits functions.

VEGF can act as a direct pro-inflammatory mediator during the pathogenesis of RA, and protect rheumatoid synoviocytes from apoptosis, which contributes to synovial hyperplasia **(**11**)**, also, the same function of VEGF-A alpha subunits which produced from TXA2 alpha subunits by the actions of mitochondrial enzymes on inflammations molecules, are protecting bone-building cells (osteoblasts) joints, and arteries from catabolic processes through reconstructing regulated anabolic cycles including mitochondrial enzymes resynthesis and reactivations to PPARs

proliferation genes and resynthesis sestrin Leu carrier activities for re-strengthen immune efficiency.

The Symptoms of Pathogenic osteoporosis is:

_Poor general health

_Low body weight,

_Estrogen deficiency,

_Poor in general, immune activities,

_Poor vision despite the correction,

_Falling,

_Full reduction in physical activity depending on the degree of osteoporosis.

The Poor in general, immune activities,and Poor in vision despite correction indicate to me that there are a poor in TXA2 alpha subunits, and thus poor in VEGF-A alpha synthesis productions and then poorly in feedback for endothelin-1 productions and poor in G-actin filaments activities and signals transmission, then the full reduction in PPARs and MAPK pathways.

The Decreasing in Muscle contraction and relaxations efficiency and the appearance of osteoporosis pathogen symptoms are so linked to each other and linked to reductions in anabolic processes and cycles means reductions in PPARs genes activities, and increasing in catabolic processes.

Stimulations to PPARs activities can be reduced by reductions in VEGF-A alpha subunits productivity, due to the reductions in TXA2 alpha subunits productions with a deficiency in AMPK proteins activities with its main MAPK pathways activities including reductions in ribosomal ATPase, or in other definitions, the main reductions in the synthesis of mitochondrial enzymes: COX2, phospholipase and in synthase enzymes will lead to reductions in TXA2 alpha, thus reduction in VEGF-A alpha subunits productions thus will lead to reductions in PPARs genes activities.

Calcified plaques were also shown to express several bone matrix proteins such as type I collagen, gla (gamma carboxyglutamate)-containing proteins, and matrix-gla protein, bone morphogenetic protein (BMP)-2 and -4, osteopontin, osteonectin, and bone sialoprotein **(**1,2,3**)**. Osteogenic cells, called calcifying vascular cells (CVCs), were identified in atherosclerotic plaques **(**1, 5**)**.

Pyrimidine and Purine nucleotide metabolic synthesis and their limit of regulations have strong relations to osteoporosis pathogens. That Purines begins with the synthesis of pyrophosphate to carbon 1 of ribose-5-phosphate, creating phosphoribosyl pyrophosphate (PRPP), where the pyrophosphate is replaced by an amine from glutamine in catabolic reactions by the effect of PRPP aminotransferase (PPAT) enzymes which are fully depending on ribosomal ATPase and G-actin ATPase activities, then the product will be 5-phosphoribosylamine (5-PRA), but in the case of a full reduction in pyrimidine nucleotides, the gamma carboxy glutamate will appear in interstitium fluid and blood looking for feedback for alpha cells and alpha subunits re-productions, and that collagen, gla (gamma carboxyglutamate has been found in the research works **(**1,2,3**)**.

Purine nucleotides have to be regulated through pyrimidine synthesis and vise versa, and that regulations are fully depending on mitochondrial enzymes that can produce synthetase enzyme which is so important for re-producing pyrimidines nucleotides from purines to resynthesis the so imp leucine amino acids which are running the most imp metabolic cycles with the help of other amino acids as tyrosine, Gly, Phe, Ser and threonine amino acids including brain activities and for resynthesis alpha subunits productions TXA2 from the activities of mitochondrial enzymes on toxicity.

Pyrimidine synthesis and their limiting in regulations in tissues cells are so important for producing availabilities of Thymine nucleotides for controlling amino acids and for re-synthesis Leu(TTA & TTG) amino acids and other branched amino acids for resynthesis sestrin-Leu carrier activities for anabolic cycles and muscles, cartilage, and joints recoverable.

The thermodynamic potential and the regulations limiting the abiotic synthesis of the common nucleobase's adenine, cytosine, guanine, and thymine, and the two monosaccharides ribose and deoxyribose and their concentrations are depending on ribosomal functions and on mitochondrial membrane activities and its production to necessary enzymes for acting on foreign molecules and toxicities including inflammation.

DNA & RNA activities are depending mainly on Thymine nucleotides synthesis and regulated with purine nucleotides, and dépending on ATPase polarizations activities, that all are necessary for anabolic reactions and catabolic processes. Where the sharp reductions in the anabolic processes reflect a sharp reduction in alpha subunits synthesis, and reductions in the stimulations to PPARs genes activities, and will reflect a sharp increase in beta subunits activities and catabolic processes, consequently, will reflect a sharp reduction in anti-inflammation cycles, and sharp increasing in toxicities and inorganic molecules in the bone marrow and their precipitation in interstitium fluid between cells and in arteries.

Is the reduction in anabolic cycles can lead to Precipitation in capillaries and arteries? Yes, reductions in anabolic processes mean reduction in PPARs genes functions means reductions in mitochondrial activities thus reductions in mitochondrial synthase, phospholipase, and COX2 enzymes mean reductions in lipid metabolism and in functioning inflammation molecules which lead to their accumulations in interstitium fluid between cells and in blood vessels, consequently will lead to isolations of some tissue cells from other tissues and preventing the G_actin signals transmission genes throughout isolated cells, that can lead to cancer problems too...

Osteoprotegerin (OPG), which is a soluble glycoprotein, and is distributed in tissues including bone-building cells (osteoblasts) are regulating both bone and vascular anabolism but under the control of the anti-inflammatory engine and its tools productions which are TXA2 alpha subunits and then it's feedback VEGF-A alpha subunits and cells. Where Osteoprotegerin is the alpha subunit that is activated by TXA2 subunits productions.

Human TXA2 subunits (which is necessary for anti-inflammations effects) receptor interact with G alpha 13 (which plays an important role in controlling cell growth), to activate intracellular signaling **(**8**)**.

The G-alpha-13 subunit (nucleotide-binding protein) is the alpha subunit that is activated by TXA2 alpha subunits productions for osteoblasts activities and vascular anabolic processes, that the activations of G alpha 13 subunits is the main.

The reduction in TXA2 subunits synthesis from the effect of mitochondrial enzymes on inflammations molecules will reflect reductions in G-alpha-13 subunit nucleotide-binding protein, and reductions in the stimulations of PPARs activities , and also, will reflect reductions in Thymine nucleotides, and reductions in Leu activities, and will reflect full reductions in most of pyrimidine nucleotides synthesis and its regulations limits controls (which done be mitochondria proper activities through active synthetase enzyme productions).

Then will lead to increasing in its beta and Gamma subunits and accumulations , where later will be characterized by the appearance of gamma carboxy glutamate which reflect the full reductions in Thymine nucleotides, and deficiency in Leu activities in interstitium fluid between tissues cells, and thus, will lead to reductions in sestrin Leu carrier tools activities, which characterized as the active tool for anabolic processes and for reactivating its proper genes signals transmitting for anabolic processes for all tissues cells including bones cells.

Synthesis of TNF-α require the presence of alpha G-protein "alpha-13 subunit of guanine" for later TGF-β synthesis, but we've to give notice that only TGF-β will cause cartilage damage in the absence & deficiency of TNF-α synthesis.

Where TGF-β subunits are the catabolic arms from alpha subunits (TNFα alpha subunits) for acting on toxicity in interstitium fluid and blood for specific alpha subunits re-productions (TXA2 alpha subunits), which will re-feedback for synthesis higher molecular alpha subunits in functions and responsibilities which is VEGF-A subunits, for later all previous steps will give the final indications for the internal anti-inflammatory tools degree of activities and efficiency.

Where, Blood-induced joint damage is fully prevented by blocking IL-1β with a monoclonal antibody or receptor antagonist, not by TNFα blockade **(**9**)**.

TNFα is secreted by the same cells in joints that synthesize IL-1β, and its increased concentration is also observed in the same elements, such as synovial fluid, synovial membrane, cartilage, and subchondral bone layer **(**10**)**.

In the cells of the joints, in regular normal cases of synovial fluid, the compositions of IL-1β have to follow active imp steps of the autocrine process under the effects of mitochondrial synthase, phospholipase, and COX2 enzymes, then COX2 respectively for acting on and functioning the toxicity of the bonded inflammation molecules through IL-1B, then by feedback process will re-produce TNFα subunits, which will stimulate the re-synthesis of their main cytokines alpha subunits TNFα, IL-6 in a limit regulations.

Results

The reductions in TXA2 alpha subunits productions through reductions in the effect of mitochondrial enzymes on inflammations molecules will reflect reductions in the feedback for VEGF-A synthesis, that will reflect a deficiency in the feedback for endothelin-1 re-synthesis and deficiency in restimulating G-actin filaments which are considered to be the necessary tools for

genes signals transmission, then will reflect deficiency and reduction in PPARs genes activities, then reduction in inflammatory system and in its necessary tools which are TXA2 and VEGF-A subunits.

Purine nucleotides have to be regulated by pyrimidine synthesis and vice versa, and that regulations are fully depending on mitochondrial active enzymes that can produce synthetase enzyme which is so important for re-producing and synthesis the Thymine pyrimidines nucleotides from purines for re-synthesis the so imp leucine amino acids which are running the most imp metabolic cycles.

The main autocrine imp steps between beta and alpha subunits productions are the so imp steps for anti-inflammations processes and are mainly controlled by mitochondrial membrane functions, by ribosomal ATPase activities and by G-actin filaments isoforms polarization activities.

Where the TNFα subunits have been observed to be secreted by cells in the joints for IL1B synthesis for functioning the inflammation molecules in the joint.

I would like to give my imp notes that all beta subunits and beta cells are for catabolic processes for functioning inflammation molecules and foreign bodies, and in normal situations are having the limit regulations effects that have to follow the autocrine imp process by the effects of mitochondrial phospholipase, COX2 and synthases enzymes for re-producing VEGF-A alpha subunits for reactivations G-actin filaments again and reactivate endothelin-1 biological molecules for ensuring and re continuing their recoveries functions through re-stimulation to MAPK pathways and PPARs activities in the proper availabilities of ribosomal ATPase activities, but alpha subunits and alpha cells are for running anabolic cycles and processes including tissue cells synthesis, and for controlling and regulating beta cells productions and activities.

Important notes Appearance of VEGF-B in rumors and interstitium fluid or blood indicate a reduction in Thymine nucleotides synthesis may due to a sever reductions in mitochondrial membrane functions and full reductions in TXA2 alpha subunits synthesis and therefore reductions in VEGF-A alpha subunits productions and lead to severe reductions in back stimulations to G-actin filaments functions and feedback for endothelin-1 resynthesis and then full reductions in the anti-inflammation engine and in its tools that can CAS many more dangerous health problems include Osteoarthritis, cancer, and cardiovascular disease and heart failure.

Many diseases are followed by a deficiency in the proper synthesis of blood cells, or deficiency in muscles functions, or poor anti-inflammatory tools VEGF-A productions through deficiency in its feedback from TXA2 alpha subunits, which produced from mitochondrial three enzymes (synthases, phospholipase, COX2).

So it is so important to realize that skeletal and vascular activities are recoveries from several diseases are fully depending on the increasing the anti-inflammatory alpha subunits keys productions (TXA2 subunits), and increasing its feedback steps for VEGF-A alpha reproductions.

TXA2 alpha subunits productions, that are activated by the effects of mitochondrial enzymes:

Phospholipase, COX2, and TXA2 synthase enzymes on inflammations molecules are so important for re-feedback for resynthesis the VEGF-A alpha subunits, which are carrying more active sites and functions for re-stimulate the endothelin_1 productions and re-stimulate G-actin filaments functions for re-complete their imp metabolic anti-inflammatory cycles, which are originated from the mitochondrial effective membrane by the productions of their active necessary synthase, phospholipase, COX2, and pyrimidine synthetase enzymes where synthetase enzyme is so important for regulating the pyrimidine nucleotides synthesis in a limit for running many imp cycles for anti-inflammations and for reactivating endothelin_1 and G_actin filaments efficiency.

It is so important to realize that skeletal and vascular activities are fully depending on the anti-inflammatory alpha subunits keys productions (TXA2 subunits), and its feedback steps for reproductions of the effective VEGF-A alpha subunits for the VEGF-B proper synthesis and productions.

References

1.Bostrom K, Watson KE, Horn S, Wortham C, Herman IM, Demer LL. “Bone morphogenetic protein expression in human atherosclerotic lesions”. J Clin Invest. 1993;91:1800–1809. [PMC free article] [PubMed] [Google Scholar] [Ref list].

2.Shanahan CM, Cary NR, Metcalfe JC, Weissberg PL. “High expression of genes for calcification-regulating proteins in human atherosclerotic plaques”. J Clin Invest. 1994;93:2393–2402. [PMC free article] [PubMed] [Google Scholar] [Ref list].

3.Dhore CR, Cleutjens JP, Lutgens E, Cleutjens KB, Geusens PP, Kitslaar PJ, Tordoir JH, Spronk HM, Vermeer C, Daemen MJ. "Differential expression of bone matrix regulatory proteins in human atherosclerotic plaques". Arterioscler Thromb Vasc Biol. 2001;21:1998–2003. [PubMed] [Google Scholar] [Ref list].

4."The link between osteoporosis and cardiovascular disease". Clin Cases Miner Bone Metab. 2008 Jan-Apr; 5(1): 19–34. PMCID: PMC2781192 PMID: 22460842 Ghada N. Farhata and Jane A. Cauleyb. Clinical Cases in Mineral and Bone Metabolism CIC Edizioni Internazionale.

5.Watson KE, Bostrom K, Ravindranath R, Lam T, Norton B, Demer LL. "TGF-beta 1 and 25-hydroxycholesterol stimulate osteoblast-like vascular cells to calcify". J Clin Invest. 1994;93:2106–2113. [PMC free article] [PubMed] [Google Scholar] [Ref list].

6.Kevin Ahern, Indira Rajagopal, and Taralyn Tan. Professor (biochemistry and biophysical) at Oregon state university. Source, Biochem FFA66. Pdf. Entire text book avaliable at, Http//Biochem. Science Oregon state. edu/content/biochemistry=free_and_easy.

7."Thermodynamic Potential for the Abiotic Synthesis of Adenine, Cytosine, Guanine, Thymine, Uracil, Ribose,and Deoxyribose in Hydrothermal Systems". Orig Life Evol Biosph (2008) 38:383–397 DOI 10.1007/s11084-008-9137-2.

8. "Coupling interaction between thromboxane A2 receptor and alpha-13 subunit of guanine nucleotide-binding protein". Chou KC. Coupling interaction between thromboxane A2 receptor and alpha-13 subunit of guanine nucleotide-binding protein. J Proteome Res. 2005;4(5):1681-1686. doi:10.1021/pr050145a.

9."IL-1β, in contrast to TNFα, is pivotal in blood-induced cartilage damage and is a potential target for therapy" Lize F. D. van Vulpen, Roger E. G. Schutgens, Katja Coeleveld, Els C. Alsema, Goris Roosendaal, Simon C. Mastbergen, Floris P. J. G. Lafeber Blood (2015) 126 (19): 2239–2246. https://doi.org/10.1182/blood-2015-03-635524.

10. "The Role of Inflammatory and Anti-Inflammatory Cytokines in the Pathogenesis of Osteoarthritis" Piotr Wojdasiewicz, Łukasz A. Poniatowski, and Dariusz Szukiewicz. online 2014 Apr 30. doi: 10.1155/2014/561459 PMCID: PMC4021678.

11."Proinflammatory Role of Vascular Endothelial Growth Factor in the Pathogenesis of Rheumatoid Arthritis: Prospects for Therapeutic Intervention" Seung-Ah Yoo,1 Seung-Ki Kwok,1 and Wan-Uk Kim1. doi:10.1371/journal.pone.0124909. Published 10 Feb 2009

12."The Gβγ-Src signaling pathway regulates TNF-induced necroptosis via control of necrosome translocation" Lisheng Li1, *, Wanze Chen1, *, Yaoji Liang1, *, Huabin Ma1, Wenjuan Li1, Zhenru Zhou1, Jie Li1, Yan Ding1 Cell Research (2014) 24:417-432. doi:10.1038/cr.2014.17; published online 11 February 2014.

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