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MOLECULAR DOCKING ELLAGIC ACID AND CALCIUM PHOSPHATE AGAINST INFLAMMATORY PROTEIN TLR2 AND TLR4 *IN SILICO*

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ABSTRACT : Inflammation is an innate immune system response to various injury stimuli, such as pathogens, injury, or metabolic stress. Inflammatory proteins that play a role in inflammatory sterile conditions (trauma) or bacterial-induced circumstances, played by TLR2 and TLR4. Ellagic acid (EA) is one of the biological molecules found in pomegranate and it has the potential anti-inflammatory. Calcium phosphate is included as the main ingredient in osteogenesis bone graft. The activity test is done to determine the potential of ellagic acid and calcium phosphate as an anti-inflammatory agent. To test the potential of ellagic acid and calcium phosphate as an anti-inflammatory agent, the biological activity was tested using the PASS Server. To understand the interaction between ellagic acid and calcium phosphate with TLR2 and TLR4, molecular docking analysis specifically used PyRx software v.0.9.5. The results of the exploration of the PubChem database of 3D structures and data SMILE shows that ellagic acid has a Pa (probability of activity) as an anti-inflammatory by 0.749. Ellagic acid and calcium phosphate tend to bind strongly to TLR2 compared to TLR4, as indicated by the results of a stronger binding affinity. Ellagic acid and calcium phosphate will remain attached to TLR but do not have a strong potential for induction of inflammation.

Key words : Calcium phosphates, ellagic acid, medicine, socket preservation, *in silico*.

INTRODUCTION

Prevention of bone resorption after tooth extraction becomes very necessary to be discussed and strongly related to the dimensions of the alveolar bone (Assistant, 2012). Residual Ridge resorption is started immediately after extraction and can result in resorption of up to 50% even in 3 months resorption will continue. After tooth extraction, alveolar ridge resorption impact on the installation of dental implants for vertical and horizontal alveolar volume should be ideally suited to the site of insertion (Horváth *et al*, 2013). Alveolar Ridge Preservation indicated to minimize the loss of ridge volume that occurs after the tooth extraction (Avila-Ortiz *et al*, 2014; Prahasanti *et al*, 2020).

The ideal bone graft materials must be biocompatible, allowing the formation of new bone or bone replacement through the osteoconductive process (Nugraha *et al*, 2019; Sari *et al*, 2020). Bovine bone xenograft (BBX) has a chemical composition and geometry of the architecture is similar to human bone and can support

new bone formation, regarded as material and as a bone graft biocompatible formation (Uzbek *et al*, 2014). Bovine bone xenograft basic materials are calcium phosphate, calcium phosphate general nature directed to the field of application Bioceramics (Canillas *et al*, 2017).

Use of BBX widely applied in alveolar bone destruction, but of several studies showed the use of alveolar bone xenograft on the vertical has very brittle nature and are not strong enough (Sheikh *et al*, 2015). Therefore, we need an innovation that is a stimulant osteoblastogenesis substance that stimulates the activity of bone graft to accelerate growth and as an anti-inflammatory that can inhibit bone resorption (Torre, 2017). Ellagic acid (EA) is one of the biological molecules found in pomegranate and have anti-inflammatory potential. Ellagic Acid detected not only in pomegranates but also in a wide variety of fruits and nuts (Usta *et al*, 2013). Ellagic acid is considered to have osteoblastogenesis stimulant is osteoinduction and has anti-inflammatory (Usta *et al*, 2013).

1 Inflammation is the innate immune system response to various stimuli that damage such as pathogens, injury, or metabolic stress. This response will work when the host immunity is not sufficient or unable to control stimulus (Antonelli and Kushner, 2017). There is a simple mechanism of occurrence of the inflammatory process which can be summarized into: (1) pattern recognition receptors on the cell surface to identify the stimulus; 2) activation of inflammatory pathways; 3) inflammatory markers released; and 4) the recruitment of inflammatory cells (Fang *et al*, 2017).

Some of PRRs can also recognize some endogenous signals are activated when there is damage to tissues or cells and associated molecular patterns recognized as damage (danger-associated molecular patterns/damps) (Chen and Nuñez, 2010; Nugraha *et al*, 2020). Endogenous stimuli can cause a sterile inflammatory response through TLR. Some purely endogenous molecules, including Heat Shock Protein (HSP), HMGB1 and uric acid, can induce the production of pro-inflammatory cytokines via TLR2 and /or TLR4 in vitro (Chen and Nuñez, 2010), Toll-like receptor (TLR) is a major regulator of innate immunity and play an integral role in the activation of the inflammatory response during infection. Moreover, TLR affects the body's response to various forms of injury (Gesueti *et al*, 2014).

The exploration of biomaterial that can support the regeneration of bone tissue is necessary needed. A synergistic combination of two ingredients needed to

inhibit the resorption of the residual ridge and keeping the alveolar ridge preservation. In this case, the approach is the *in silico* molecular docking to determine the effects of ellagic acid and calcium phosphate to proteins inflammatory TLR 2 and TLR 4.

MATERIALS AND METHODS

To test the potential of ellagic acid and calcium phosphate as an anti-inflammatory agent, tested the biological activity using the PASS Server. To understand the interaction between ellagic acid and calcium phosphate with TLR2 and TLR4, an analysis of specific molecular docking using PyRx software v.0.9.5.

RESULTS

TLR pathway exploration results (toll-like receptor) which may occur in visualizing through KEGG database (map04620) <https://www.genome.jp/kegg/pathway.html>. Inflammatory proteins that play a role in inflammatory sterile conditions (trauma) or bacterial, played by TLR2 and TLR4 [13]. This is supported by data KEGG Pathway indicating that TLR2 and TLR4 have direct interaction with bacterial lipoproteins and the location of the protein is in the cell membrane (Fig. 1, red box). So that TLR2 and TLR4 become a target protein for testing the effectiveness of ellagic acid and calcium phosphate.

Exploration results from the PubChem database of 3D structures and data SMILE are shown in Figure 2. shows that ellagic acid has a Pa (probability of activity) as an anti-inflammatory by 0.749. Scores above 0.7

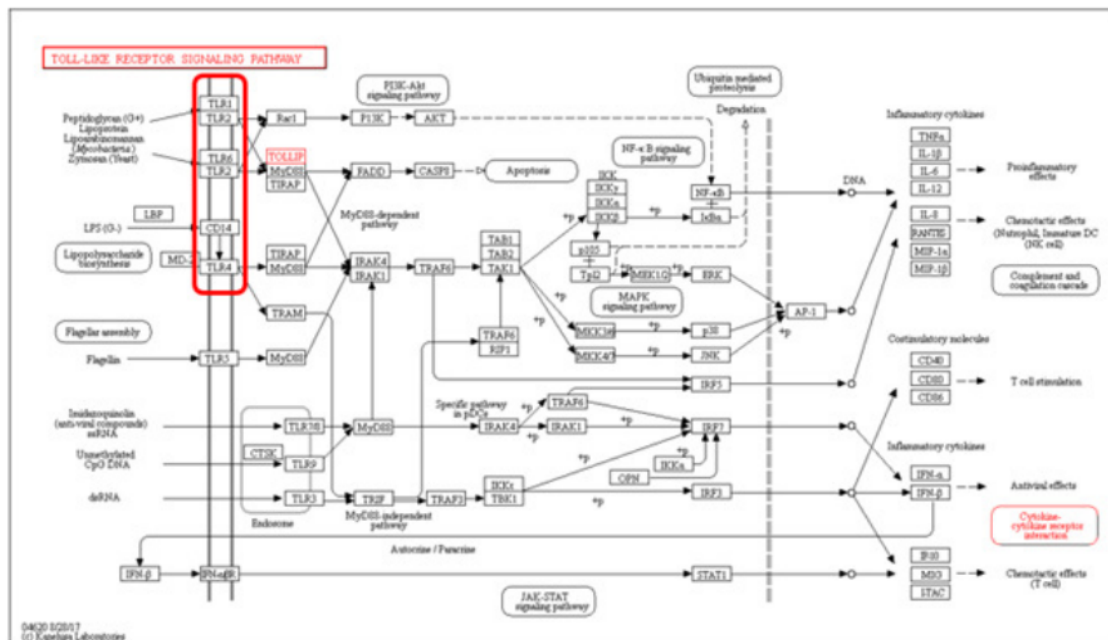


Fig. 1 : Location of proteins located in the cell membrane (red box) (Shen, Kreisel and Goldstein, 2013).

2
Molecular docking ellagic acid and calcium phosphate against inflammatory protein TLR2 and TLR4 *in silico*

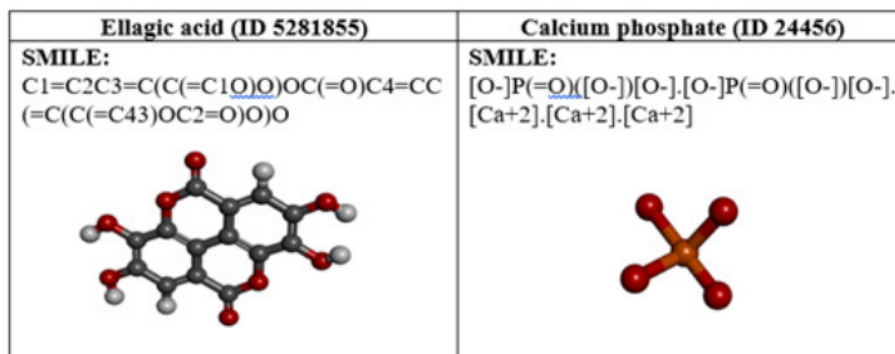


Fig. 2 : 3D structure, SMILE ellagic acid and calcium phosphate.



Fig. 3 : Interaction of molecular docking between the TLR4-MD2 (wheat; cartoon) with LP5 (control) (red; line), calcium phosphate (green; line) and ellagic acid (cyan; line)

Table 1 : Results of docking per molecule to the receptor TLR.

Receptor	Ligan	Binding Affinity (kcal/mol)
TLR4-MD2	LP5	-47.07
	Ellagic acid	-35.82
	Calcium Phosphate	-23.99
TLR2/TLR6	PSX	-84.55
	Ellagic acid	-50.12
	Calcium Phosphate	-21.27

Table 2 : Results docking combination of ellagic acid and calcium phosphate to the receptor TLR.

Receptor	Ligan	Binding Affinity (kcal/mol)
TLR4-MD2	Ellagic acid dan Calcium Phosphate	-21.28
TLR2/TLR6	Ellagic acid dan Calcium Phosphate	-22.04

indicate the accuracy of the prediction server. Information and SMILE 3D structure of ellagic acid (ID 5281855) and calcium phosphate (ID 24456) were obtained from the NCBI PubChem database (<https://pubchem.ncbi.nlm.nih.gov/>).

2
To understand the interaction between ellagic acid and calcium phosphate with TLR2 and TLR4, an analysis of specific molecular docking using PyRx software v.0.9.5. TLR4-MD2 complex (PDB ID 3VQ1) and TLR2 / TLR6 (PDB ID 3A79) as a receptor obtained from the

PDB database (<http://www.rcsb.org/>). Molecular docking was conducted using specific docking, which is directed at the active site control using Patchdock (<https://bioinfo3d.cs.tau.ac.il/PatchDock/>).

Results signaling through TLR2 would happen if he forms a heterodimer complex with TLR1 or TLR6 (Jin *et al*, 2007). Complex TLR2 / TLR1 or TLR2 / TLR6 not show differences in signaling pathways. Some TLR2 complex that has been modelled always form a heterodimer with TLR1 or TLR6 (Farhat *et al*, 2008).

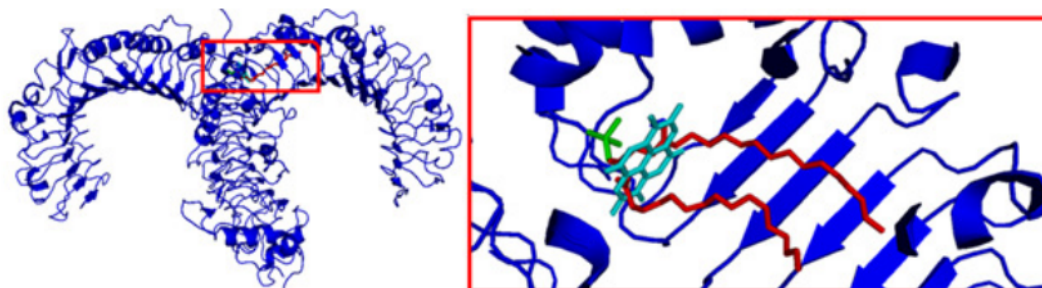


Fig. 4 : The interaction of molecular docking between TLR2 / TLR6 (blue; cartoon) with PXS (control) (red; line), calcium phosphate (green; line) and ellagic acid (cyan; line).

Table 3 : The same amino acid to the receptor TLR 4.

Receptor	Ligan	Ikatan hidrogen	Ikatan hidrofobik
TLR4-MD2 (3VQ1)	LP5	-	Phe126, Ile80, Ile153 , Cys133 , Phe151 , Ile52, Ser48, Val61, Ile117, Phe119, Ile124, Phe121
	Ellagic acid	-	Ile52, Ile153 , Val61, Ile46, Leu78, Phe76, Ala135, Phe151 , Cys133 , Ile32
	Calcium Phosphate	Unable to analyze the bond	

Table 4 : The same amino acid to the receptor TLR 2.

TLR2/TLR6 (3A79)	PSX	Phe349	Phe325 , Lys347 , Ser346 , Ile319, Leu317, Phe284, Thr262, Leu335, Leu261, Phe266, Leu269, Leu270, Phe355, Leu328 , Val351, Leu350 , Pro352
	Ellagic acid	-	Asp327, Leu328 , Tyr326, Pro352 , Phe325 , Phe349, Lys347 , Phe322, Ser346 , Val348, Phe355, Leu350
	Calcium Phosphate	Unable to analyze the bond	

Molecular docking analysis showed that TLR4 has the strongest ties with LP5 as a control, followed by ellagic acid and calcium phosphate with a score of -47.07 respectively; -35.82 and -23.99 kcal/mol. The same results are also shown in the interaction of TLR2/TLR6 with PSX as a control with the highest affinity binding -84.55 kcal/mol. Furthermore, ellagic acid and calcium phosphate with a score of -50.12 and -21.27 kcal/mol. Ellagic acid and calcium phosphate attached to the same site to control, visualize with Figs. 3 and 4.

The data show that ellagic acid and calcium phosphate tends to bind strongly to TLR2 compared to TLR4, as

indicated by the results of a stronger binding affinity. This may occur because when the small molecules bind to TLR4, then he must first bind with a special pocket called MD2 (Fig. 3). Meanwhile, if viewed from the bond between TLR2 with small molecules is bonding directly to TLR4 (without intermediaries) (Fig. 4).

Based on the literature, TLR4 induces the formation of Th1-inducing cytokine IL12, p70 and interferon-gamma inducible protein chemokine IP-10, whereas stimulation of TLR2 was not able to induce gamma interferon IL12 P70 and IP10, but produce IL12 inhibitor p40 homodimer that makes developing good Th2 (Re and Strominger,

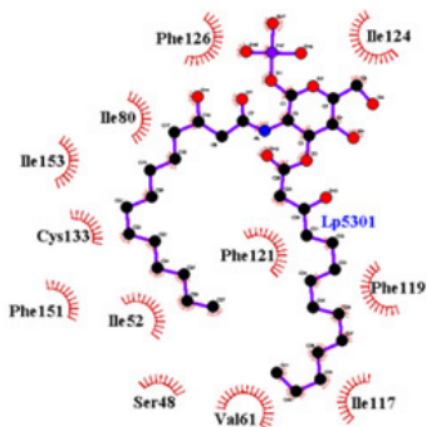


Fig. 5 : The amino acids involved in the TLR4-MD2 with LP5 (control).

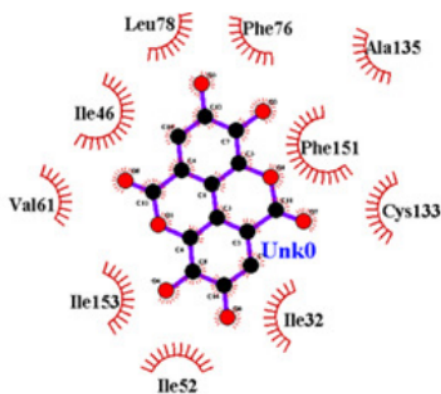


Fig. 6 : The amino acids involved in the TLR4-MD2 with Ellagic acid.

2001). Also, the potential of ellagic acid and calcium phosphate to induce TLR2 or TLR4 was not as good as LPS control (PSX or LP5). Thus, there is a potential that ellagic acid and calcium phosphate did not induce massive inflammation.

Results showed that the combination of ellagic acid and calcium phosphate to show binding affinity TLR receptors is decreased compared to the docking per molecule (Tables 1 and 2). This strengthens the case that the second component of TSB will remain attached to TLR but does not have a strong potential for the induction of inflammation. Also, the adhesion between the ellagic acid phosphate and calcium differently, if the comparison between the individual and combined docking.

The interaction of molecular docking showed that the TLR-ellagic acid complex has several sides of the same attachment, shown with the amino acid marked with highlights. Amino acids that have amino acid similarity highlights indicate that contribute to the control and treatment.

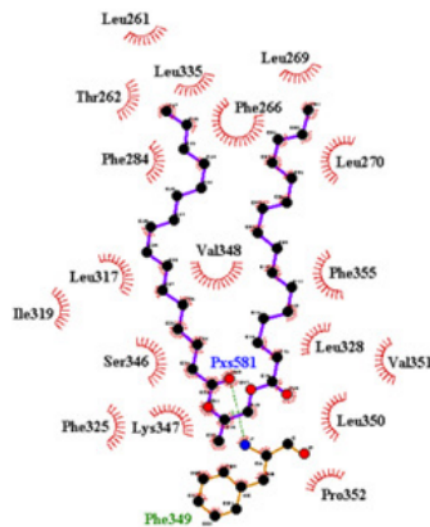


Fig. 7 : The amino acids involved in the TLR2 / TLR6 with PXS (control).

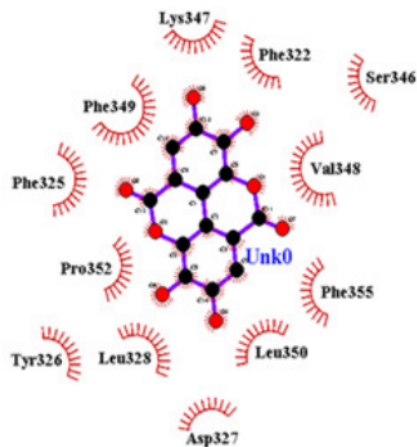


Fig. 8 : Amino acids are involved between the TLR2 / TLR6 with ellagic acid.

DISCUSSION

Based on the results that ellagic acid has a probability of activity (Pa). It has the potential to lower the anti-inflammatory properties. According to the Lee study, suggesting that blockade of TLR signaling pathway is ellagic acid therapeutic strategy for the treatment of liver disease (Lee *et al*, 2014). Previous studies have demonstrated that TLR2 and TLR4 involved in the mechanisms of inflammation in hepatitis (Soares *et al*, 2012). TLR is one of the principle involved in the innate immune receptors as mediators of certain sterile inflammatory conditions. In both models, TLR2 and TLR4 was shown to signal through the adapter MyD88 signaling to contribute to inflammation (Shen *et al*, 2013).

Ellagic acid is proven to be an effective anti-

inflammatory agents in rats induced foot edema carrageenan, with a long onset and duration of action (Corbett *et al*, 2010), Ellagic acid is a COX inhibitor, is a strong anti-inflammatory and anti-nociceptive agents observed through inhibition of cyclooxygenase (COX) (Banihani *et al*, 2015). Ellagic acid can also reduce inflammation in rat colon carcinogenesis (Umesalma and Sudhandiran, 2010), Mediates anti-rheumatic effect via down-regulation of proinflammatory cytokines and upregulation of anti-inflammatory cytokines (Allam *et al*, 2016).

Results docking per molecule to the receptor TLR showed that Ellagic acid and calcium phosphate tends to bind strongly to TLR2 compared to TLR4. Molecular docking analysis showed that TLR4 has the strongest ties with LP5 as a control, followed by ellagic acid and calcium phosphate. This may occur because when the small molecules bind to TLR4, then he must first bind with a special pocket called MD2. Wang appropriate research, Toll-like receptor 4 (TLR4) / myeloid differentiation factor 2 (MD-2) recognizes lipopolysaccharide (LPS) in Gram-negative bacteria to induce an innate immune response. Neoseptin, peptidomimetic chemically synthesized that binds to and activates the mouse TLR4 (mTLR4) / MD-2 independent of LPS (Ying Wang *et al*, 2016). Previous studies have shown that MD-2 is associated with TLR4 on the cell surface and allows TLR4 to respond to LPS. TLR2 without MD-2 does not respond to LPS, ReLPS, and lipid a free pure protein. MD-2 is physically associated with TLR4 and TLR2, but the relationship with TLR2 weaker than with TLR4 (Dziarski and Gupta, 2000).

The bond between TLR2 with a small molecule is a direct bond to TLR4. New inhibitors TLR1 / TLR2 obtained with the cell-based screening of small molecule libraries of NCI-2 Diversity comprising of 1363 compounds. Subsequent research/ must identify small molecules to bind TLR1 / TLR2 (Cheng *et al*, 2012), TLR4 activation or inhibition by small molecules in the next few years will be a new therapy. TLR4 also recognize endogenous molecules, called damp, released by tissue injury and necrotic cells. TLR4 is the key receptor in which the stimuli are not infectious and infectious met to trigger a pro-inflammatory response (Zaffaroni and Peri, 2018),

Results docking combination of ellagic acid and calcium phosphate to the receptor TLR showed that the combination of ellagic acid and calcium phosphate to show binding affinity TLR receptors is decreased compared to the docking per molecule. This is because the molecules have different receptors. Ellagic acid if in

a separate docking to the receptor TLR showed a high binding affinity than if the docking combination with calcium phosphate. In the study of Wang explained that the Wnt and Notch signaling pathway plays an important role in the differentiation of osteogenic which is enhanced by calcium phosphate, but the mechanism of synergistic effects needs to be explored further (Wang *et al*, 2019),

Calcium phosphate is among the best substitute for bone graft for rapid bone formation on the surface, and the possibility of bone healing in a year. Several reasons explain the excess calcium phosphate: (i) as a biological apatite as calcium and phosphate ions are present in large amounts in the human body; (ii) Calcium phosphate is absorbed by the cell-mediated process, ensuring not only the material simultaneously absorption and bone formation, but also there is no problem of biocompatibility; (iii) calcium and phosphate ions have a strong effect directly on bone cells, with particular phosphate ions triggers a response considered osteoinductive (Habraken *et al*, 2016), The combination of both materials in silico prediction is expected to help the researchers to continue research *in vivo* tests or *in vitro* tests.

CONCLUSION

Ellagic acid and calcium phosphate will remain attached to TLR, but do not have a strong potential for induction of inflammation *in silico*.

Conflict of interest

The authors declare no conflicts of interest.

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REFERENCES

- Allam G, Mahdi E A, Alzahrani A M and Abuelsaad A S (2016) Ellagic acid alleviates adjuvant induced arthritis by modulation of pro- and antiinflammatory cytokines. *Central Europ. J. Immunol.* **41**(4), 339–349. Retrieved from <https://doi.org/10.5114/ceji.2016.65132>
- Antonelli M and Kushner I (2017) It's time to redefine inflammation. *FASEB J.* **31**(5), 1787–1791. Retrieved from <https://doi.org/10.1096/fj.201601326R>
- Avila-Ortiz G, Elangovan S, Kramer K W O, Blanchette D and Dawson D V (2014) Effect of alveolar ridge preservation after tooth extraction: A systematic review and meta-analysis. *J. Dental Res.* **93**(10), 950–958. Retrieved from <https://doi.org/10.1177/0022034514541127>
- Banihani S, Swedan S, Alguraan Z, Cheng I, Witte J S, McClure L A and Khan F (2015) Degradation Kinetics of Bioactive Compounds and Antioxidant Activity of Pom: 'Start Your Research'. *Food Chem.* **18**(3), 575–584. Retrieved from <https://doi.org/10.1016/j.foodchem.2014.12.003>

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- Canillas M, Pena P, De Aza A H and Rodríguez M A (2017) Calcium phosphates for biomedical applications. *Boletín de La Sociedad Española de Cerámica y Vidrio* **56**(3), 91–112. Retrieved from <https://doi.org/10.1016/j.bsecv.2017.05.001>
- Chen G Y and Nuñez G (2010) Sterile inflammation: Sensing and reacting to damage. *Nature Reviews Immunol.* **10**(12), 826–837. Retrieved from <https://doi.org/10.1038/nri2873>
- Cheng K, Wang X, Zhang S and Yin H (2012) Discovery of small-molecule inhibitors of the TLR1/TLR2 complex. *Angewandte Chemie* **51**(49), 12246–12249. Retrieved from <https://doi.org/10.1002/anie.201204910>
- Corbett S, Daniel J, Drayton R, Field M, Steinhardt R and Garrett N (2010) Evaluation of the anti-inflammatory effects of ellagic acid. *J. Perianesthesia Nursing* **25**(4), 214–220. Retrieved from <https://doi.org/10.1016/j.jopan.2010.05.011>
- Dziarski R and Gupta D (2000) Role of MD-2 in TLR2- and TLR4-mediated recognition of Gram-negative and Gram-positive bacteria and activation of chemokine genes. *J. Endotoxin Research.* Retrieved from <https://doi.org/10.1179/096805100101532243>
- Fang J, Zhao L, Deng J, Chen L, Li Y, Cui H and Deng H (2017) Inflammatory responses and inflammation-associated diseases in organs. *Oncotarget.* **9**(6), 7204–7218. Retrieved from <https://doi.org/10.18632/oncotarget.23208>
- Farhat K, Riekenberg S, Heine H, Debarry J, Lang R, Mages J and Ulmer A J (2008) Heterodimerization of TLR2 with TLR1 or TLR6 expands the ligand spectrum but does not lead to differential signaling. *J. Leukocyte Biol.* **83**(3), 692–701. Retrieved from <https://doi.org/10.1189/jlb.0807586>
- Gesuete R, Kohama S G and Stenzel-Poore M P (2014) Toll-like receptors and ischemic brain injury. *J. Neuropathol. and Exp. Neurol.* **73**(5), 378–386. Retrieved from <https://doi.org/10.1097/NEN.0000000000000068>
- Habraken W, Habibovic P, Eppele M and Böhner M (2016) Calcium phosphates in biomedical applications: Materials for the future? *Materials Today* **19**(2), 69–87. Retrieved from <https://doi.org/10.1016/j.mattod.2015.10.008>
- Horváth A, Mardas N, Mezzomo L A, Needleman I G and Donos N (2013) Alveolar ridge preservation. A systematic review. *Clinical Oral Investigations* **17**(2), 341–363. Retrieved from <https://doi.org/10.1007/s00784-012-0758-5>
- Jin H, Xu J and Wen Z (2007) Migratory path of definitive hematopoietic stem/progenitor cells during zebrafish development. *Blood* **109**(12), 5208–5214. Retrieved from <https://doi.org/10.1182/blood-2007-01-069005>
- Lee J H, Won J H, Choi J M, Cha H H, Jang Y J, Park S and Kim D K (2014) Protective effect of ellagic acid on concanavalin A-induced hepatitis via TLR- and MAPK/NF- κ B signaling pathways. *J. Agricult. and Food Chem.* Retrieved from <https://doi.org/10.1021/jf503188c>
- Nugraha A P, Narmada I B, Sitasari P I, Inayati F, Wira R and Triwardhani A (2020) High mobility group box 1 and heat shock protein-70 expression post (-)-epigallocatechin-3-gallate in East Java green tea methanolic extract administration during orthodontic tooth movement in wistar rats. *Pesqui Bras Odontopediatria Clin Integr.* **20**, e5347, 1-6.
- Nugraha A P, Rezki F, Putra K G, Narmada I B, Ernawati D S and Rantam F A (2019) Triad Tissue Engineering: Gingival Mesenchymal Stem Cells, Platelet Rich Fibrin and Hydroxyapatite Scaffold to ameliorate Relapse Post Orthodontic Treatment. *Biochem. Cell. Arch.* **19**(2), 3689–3693.
- Prahasanti C, Nugraha A P, Saskianti T, Suardita K, Riawan W and Ernawati D S (2020) Exfoliated Human Deciduous Tooth Stem Cells Incorporating Carbonate Apatite Scaffold Enhance BMP-2, BMP-7 and Attenuate MMP-8 Expression During Initial Alveolar Bone Remodeling in Wistar Rats (*Rattus norvegicus*). *Clinical, Cosmetic and Investigational Dentistry* **12**, 79–85.
- Re F and Strominger J L (2001) Toll-like Receptor 2 (TLR2) and TLR4 Differentially Activate Human Dendritic Cells. *J. Biol. Chem.* **276**(40), 37692–37699. Retrieved from <https://doi.org/10.1074/jbc.M105927200>
- Sari D S, Maduratna E, Ferdiansyah, Latief F D E, Satuman, Nugraha A P, Sudiana K and Rantam FA (2019) Osteogenic Differentiation and Biocompatibility of Bovine Teeth Scaffold with Rat Adipose-derived Mesenchymal Stem Cells. *Eur. J. Dent.* **13**(2), 206–212.
- Sheikh Z, Sima C and Glogauer M (2015) Bone replacement materials and techniques used for achieving vertical alveolar bone augmentation. *Materials* **8**(6), 2953–2993. Retrieved from <https://doi.org/10.3390/ma8062953>
- Shen H, Kreisel D and Goldstein D R (2013) Processes of Sterile Inflammation. *The J. Immunol.* **191**(6), 2857–2863. Retrieved from <https://doi.org/10.4049/jimmunol.1301539>
- Soares J B, Pimentel-Nunes P, Afonso L, Rolanda C, Lopes P, Roncon-Albuquerque R and Leite-Moreira A F (2012) Increased hepatic expression of TLR2 and TLR4 in the hepatic inflammation-fibrosis-carcinoma sequence. *Innate Immunity* **18**(5), 700–708. Retrieved from <https://doi.org/10.1177/1753425912436762>
- Torre E (2017) Molecular signaling mechanisms behind polyphenol-induced bone anabolism. *Phytochemistry Reviews* **16**(6), 1183–1226. Retrieved from <https://doi.org/10.1007/s11101-017-9529-x>
- Umesalma S and Sudhandiran G (2010) Differential inhibitory effects of the polyphenol ellagic acid on inflammatory mediators NF- κ B, iNOS, COX-2, TNF- α and IL-6 in 1,2-dimethylhydrazine-induced rat colon carcinogenesis. *Basic and Clinical Pharmacology and Toxicol.* **107**(2), 650–655. Retrieved from <https://doi.org/10.1111/j.1742-7843.2010.00565.x>
- Usta C, Ozdemir S, Schiariti M and Puddu P E (2013) The pharmacological use of ellagic acid-rich pomegranate fruit. *Int. J. Food Sciences and Nutrition* **64**(7), 907–913. Retrieved from <https://doi.org/10.3109/09637486.2013.798268>
- Uzbek U H, Rahman S A B, Alam M K and Gillani S W (2014) Bone forming potential of an-organic bovine bone graft: A cone beam CT study. *J. Clin. and Diagnostic Research* **8**(12), ZC73–ZC76. Retrieved from <https://doi.org/10.7860/JCDR/2014/8557.5352>
- Wang Y, Su L, Morin M D, Jones B T, Whitby L R, Surakattula M M R P and Beutler B (2016) TLR4/MD-2 activation by a synthetic agonist with no similarity to LPS. *Proceedings of the National Academy of Sciences of the United States of America* **113**(7), E884–E893. Retrieved from <https://doi.org/10.1073/pnas.1525639113>
- Wang Y, Pan J, Zhang Y, Li X, Zhang Z, Wang P and Li J (2019) Wnt and Notch signaling pathways in calcium phosphate-enhanced osteogenic differentiation: A pilot study. *J. Biomedical Materials Research - Part B Applied Biomaterials* **107**(1), 149–160. Retrieved from <https://doi.org/10.1002/jbmb.34105>
- Zaffaroni L and Peri F (2018) Recent advances on Toll-like receptor 4 modulation: New therapeutic perspectives. *Future Medicinal Chem.* **10**(4), 461–476. Retrieved from <https://doi.org/10.4155/fmc-2017-0172>

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