In silico docking study of salicylic acid on gene expression during wound healing

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ABSTRACT

Wound healing is an intricate process characterized by hemostasis, inflammation, proliferation and remodeling. Upon injury to the skin, a set of complex biochemical events takes place simultaneously to repair the damage. Tissue damage also triggers a robust influx of inflammatory leukocytes to the wound site that play key roles in clearing the wound from invading microbes and also release signals that may be detrimental to repair and lead to fibrosis. In the present *in silico* investigation, an attempt was made to identify the genes (C5a anaphylatoxin chemtactic receptor 1, Growth regulated alpha protein and A rachidonate-5-lipoxygenase activating protein) which are expressed throughout the wound healing and their interaction with the salicylic acid extracted from the floral extract of *Tagetes erecta Linn*. The identified proteins were evaluated for stability by Ramachandran Plot and the three dimensional structures were validated using PROCHEK server. The inhibiting susceptibility of salicylic acid was evaluated using GOLD software. The results were evaluated based on the binding compatibility (ie) Docked energy in K Cal/ Mol (fitness) and conformed based on the number of hydrogen bonds formed and the bond distance between atomic co-ordinates. The maximum fitness score of 31.29 was recorded against C5a anaphylatoxin chemtactic receptor 1 followed by Arachidonate -5-lipoxygenase activating protein (28.57) and the growth regulated alpha protein scored the least of 25.82.

KEYWORDS : *In silico*, validated, expressed, docking, score.

1. INTRODUCTION

Skin is well recognized as an important somatic mirror of one's emotion and a site for the discharge of one's anxieties (Sobanko and Alster, 2009) and a complex array of reparative tissue mechanism occurs in the skin after epithelial healing reponses that result in inadequate restoration of thye cutaneous surface (Meenakshi et al., 2005). Hypertrophic scars and keloids are common problems after injury and cause functional and cosmetic deformities (Devika and Nazareth, 2011). Physiological responses after thermal injury includes changes in cellular protection mechanism (Jackson, 1993), local and systemic inflammation (Marcus, 2002) and reperfusion injury which abrupt cellular energy depletion and cell damage (Mallouk et al., 1999; Nesic et al., 2002). Inflammation mediated delayed cell death occurs at the wound borders and in surrounding tissues, increases accumulation of macrophages and fibroblasts which leads to extracellular matrix deposition, angioneogenesis to form granulation tissue (Gibans and Heimbach, 2000 : Lawrence, 1998). The focus of the present study is to identify the genes which are expressed throughout the process of wound healing anf their interaction with the sacylic acid ligand using GOLD software.

2. MATERIALS AND METHODS

2.1. Active Site Prediction: The binding site of three proteins namely C5a anaphylatoxin chemtactic receptor1, Growth regulated alpha protein and Arachidonate 5-lipoxygenase activating protein were determined using CASTp.

2.2. Protein Stability: The stability of the three proteins was determined using Ramachandran Plot.

2.3. Ligand Preparation: The 2D structures of flavonoid was drawn using ACD chemsketch and the structures were then converted to their 3D geometrics, optimized and saved in " MDL mol file" format.

2.4. Docking Simulation: Automated docking studies were performed using the genetic algorithm GOLD (Version 3.2 CCDC, Cambridge, U.K)

3. RESULTS AND DISCUSSION

The sequence of burn wound healing virtually starts with the injury itself. Inflammation related proteins like C5a anaphylatoxic chemotactic receptor1, Growth related alpha protein and Arachidonate 5 lipoxygenase activating proteins were predicted using Muster and Modeller 9.10 and the stability of the proteins were verified with Ramachandran Plot (Table.1).

Table 1 : Validation of selected target proteins

Protein	Predicted Method	Stability of Protein in Ramachandran Plot
C5a anaphylatoxin chemostactic receptor 1	Muster	100
Growth regulated alpha protein	Modeller 9.10	100
Arachidonate 5-lipoxy genase activating protein	Modeller 9.10	100

Table.2.Protein – Ligand interactions using GOLD

Ligand Name	C5a anaphylatoxin chemotactic receptor 1			
	Atom in protein	Atom in Ligand	H – Bond distance	Score
Salicylic acid	ASP37:OD2 HIS100:NE2	O10 O9	2.566 3.055	31.29
Growth regulated alpha protein				
Salicylic acid	MET100:N ILE96:O	O7 O10	2.817 2.355	25.82
Arachidonate 5- lipoxygenase activating protein				
Salicylic acid	THR5:O	O10	2.889	28.57

The three dimensional structures of the receptors has been predicted and was subjected to validation using PROCHEK server. The Ramachandran Plot showed 100% of amino acid in the allowed region which indicated the overall stable conformation of the protein structure. The inhibiting susceptibility of salicylic acid was evaluated using GOLD software. The results were evaluated based on the binding compatibility (ie) Docked energy in K Cal/ Mol (Fitness). This was conformed based on the number of hydrogen bonds formed and bond distance between atomic Co-ordinates of the active sites of C5a anaphylatoxin chemotactic receptor1 (Fig.1), Growth regulated alpha protein (Fig.2) and Arachidonate 5- lipoxygenase activating protein (Fig.3) and conformation results are tabulated in the Table.2. Least interaction score of salicylic acid was obtained against C5a anaphylatoxin chemotactic receptor 1 with the fitness score of 31.29 where the interaction was stabilized by 3 hydrogen bonds. The Second maximum fitness of 28.57 was recorded against Arachidonate 5-lipoxygenase activating protein with salicylic acid followed by growth regulated alpha protein with 25.82 score. From the above *in silico* investigation salicylic acid compound showed effective fitness against the wound healing processes.

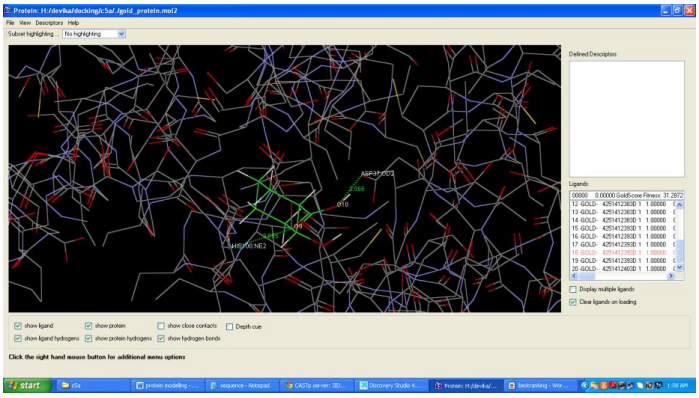

Fig. 1 Interaction of C5a anaphylatoxin chemotactic receptor 1 with salicylic acid

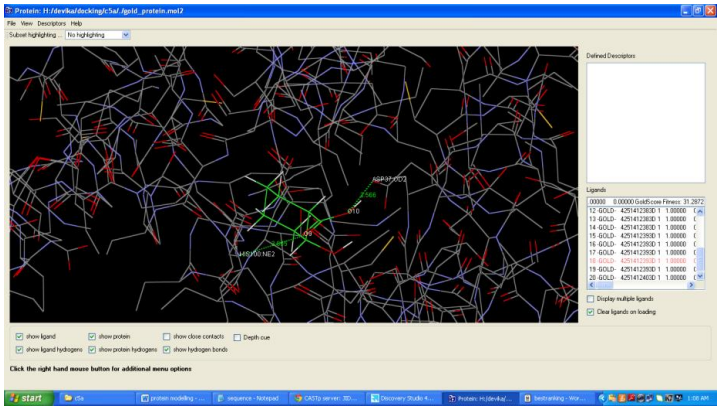

Fig.2 Interaction of Growth-regulated alpha protein with salicylic acid

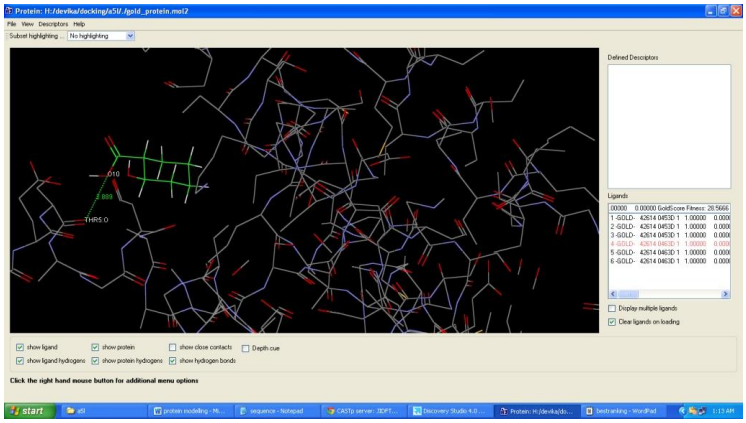

Fig.3 Interaction of Arachidonate 5-lipoxygenase-activating protein with salicylic acid

4. CONCLUSION

In silico investigation towards the protein ligand interactions proved that the bioactive compound salicylic acid has the maximum fitness score with GOLD docking and it also paves a way of conformation that salicylic acid may have the best anti-inflammatory role in wound healing.

REFERENCES

Devika R and Nazareth Arockiamary S, Aetiology of Keloids- An overview Res in Biotech., 2 (6), 2011, 37-43.
Gibans NS and Heimbach DM. Current status of burn wound pathophysiology. Clin Plast Surg., 27, 2000, 11-22.
Jackson D, The diagnosis of the depth of burning. J Br Surg., 40, 1993, 585-596.
Lawrence WT, Physiology of the acute wound. Clin Plast Surg., 25, 1998, 321-340.
Mallouk Y, Vayssier Taussat M, Bonventre JV and POlla RS. Heat shock protein 70 and ATP as partners in cell homestasis. Int J Mol Med., 4, 1999, 463-474.
Marcus spies, Mohan RK, Dasu, Nenad svrakie, Olivera Nesic, Robert E barrow, Regino Perez Polo J and David N Herndon. Gene expression analysis in burn wounds of rats. Am J Physiol Regul Integr Comp Physiol., 283, 2002, R918-R930.
Meenakshi V, Jayaraman KM, Ramakrishnan and Balu M. Keloids and Hypertrophic scars A review. Int J Plast Surg., 38, 2005, 175-9.
Nesic O, Syrakie NM, Xu GY, MsAdoo D, Westlund KN, Hulsebosch CE, Ye Z, Galante A, Soteropouloss P, Tolis P, Young W, Hart RP and Perez polo JR, DNA micro array analysis of the confused spinal cord: Effect of NMDA inhibition. J Neurosci Res., 68, 2002, 406-423.
Sobanko JF and Alster TS, Laser treatment for scars and wounds, Glornale Italian di dermatologia e vernereologia, 144(5), 2009, 583-93.