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Original Research Article

Investigation of the anti-aging properties of allicin from *Allium sativum* L bulb extracts by a reverse docking approach

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Abstract

Purpose: *To analyze the potential of Allium sativum bulb extract in curing premature aging using reverse docking method.*
Methods: *Ligand samples were retrieved from a PubChem database, allicin CID 65036, epigallocatechin gallate CID 65064, and pedunculagin CID 442688. To improve reliability, protein targets were predicted using three web services (PharmMapper, SuperPred and Swiss Target Prediction). Molecular docking was conducted to predict the interaction between Allium sativum bioactive as ligands and leukocyte elastase as protein target using PyRx 0.8 software. To be sure of the drug potential of the bioactive, DruLiTo software was used for the evaluation. Visualization and interaction analysis were performed by PyMol and Ligplus software.*
Results: *The results showed that allicin has the highest potential as a candidate for premature-aging treatment, as evidenced by the highest binding affinity (-8.7 kcal/mol) to leukocyte elatase compared with epigallocatechin (-7.2 kcal/mol) and pedunculagin (-7.8 kcal/mol). Allicin acted as a leukocyte elatase inhibitor, with its binding stability facilitated by hydrogen bonding and hydrophobic interactions.*
Conclusion: *Allicin has a potential as a leukocyte elastase inhibitor based on its binding affinity and intermolecular interactions. Thus, allicin is a potential anti-aging drug candidate based on Lipinski's rule.*

Keywords: *Anti-aging, Allicin, Leukocyte elastase inhibitor, Protein target analysis, Binding affinity, Reverse docking*

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INTRODUCTION

The presence of free radicals can cause various diseases including premature aging [1]. Skin aging is a complex evitable process for all living organisms [2,3]. There are several skincare cosmetics that can treat premature aging, but many of them can have adverse reactions including allergic contact dermatitis, irritant contact dermatitis, as well as phototoxic and photoallergic reactions [2,4]. To avoid some of these adverse effects, recently, anti-aging research has focused on scientific validation of natural products, often used throughout history for their medicinal properties [5]. Natural products like fruit, vegetables, spices provide antioxidant bioactive compounds and have been

widely used as a cosmetics historically and to the present day in some societies [6]

Garlic (*Allium sativum* L.) is a well-known spices used in daily life especially for foodstuff and medicines in many cultures. Many people used garlic to prevent and treat several diseases [7,8]. Treatments using this herbal medicine need further verification to understand the mechanism of its activity. One useful bioinformatic-based method to reveal and understand the potential of *A. sativum* is the reverse docking method. This technique is popular for predicting the interaction between active compound and protein target.

Most of the active compounds from *A. sativum* have phenolic groups. Allicin is the main bioactive component [11,12]. Allicin has a strong antioxidant activity as a protective compounds against free radical damage [13-15]. It has been observed experimentally that allicin can cure premature aging. [16,17]. This research aimed to analyze the potential of allicin in *A. sativum* in curing premature aging by the reverse docking method.

EXPERIMENTAL

Ligand preparation

The chemical 3D structure and SMILES of ligand (allicin CID 65036, epigallocatechin gallate CID 65064, and pedunculagin CID 442688) were retrieved from the PubChem compound database (https://pubchem.ncbi.nlm.nih.gov/).

Target selection and molecular docking analysis

Ligands structure were analyzed using Pharmmapper (http://lilab.ecust.edu.cn) to identify target protein based on pharmacophore mapping approach [10] and ligands SMILES format were analysed using SuperPred (http://prediction.charite.de/) for identifying the target protein based on combination of physicochemical properties and similarity [18]. The specific protein target of *A. sativum* ligands was predicted using Swiss Target Prediction (http://swisstargetprediction.ch) [19]. Molecular docking of allicin, epigallocatechin and pedunculagin analysis was done using Pyrx 0.8. We used reference inhibitor reference for validating the docking results. The potential inhibitors were screened using DruLito based on Lipinski's rule [20]**.** The molecular docking result was analysed by LigPlus V.2.0 to understand the details of the interactions between ligands and the protein. All visualizations of Biomolecules were conducted by PyMol Software.

RESULTS

The results of target selection showed that allicin interacted with leukocyte elastase (Supp. Data 1). The 3D structure of leukocyte elastase was collected from the Protein Data Bank database (www.rcsb.org/) (ID number 2Z7F) and visualized using PyMOL software (Figure 1). Molecular docking modelling showed that allicin had the highest affinity with leukocyte elastase (-8,7 kcal/mol) compared with epigallocatechin gallate (-7,2 kcal/mol), and pedunculagin (-7,8 kcal/mol).

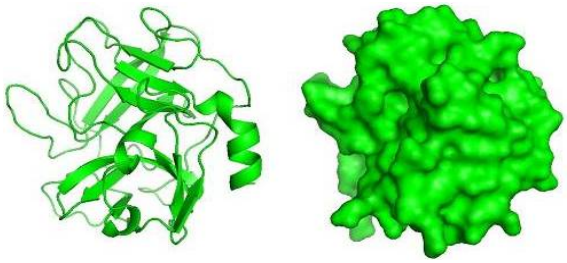

Figure 1: 3D structure of leukocyte elastase visualyzed using PyMOL software

Moreover, molecular interaction showed that allicin was predicted to interact with leukocyte elastase via hydrogen bonds with of Gly 193, Ser 195 and via hydrophobic interactions with Val 190, Cys 191, Phe 192, Ser 214, Phe 215, and Val 216 (Figure 2). Epigallocatechin gallate was predicted to interact with leukocyte elastase via His 57, Asn 61, Tyr 94, Asp 102, Ser 195, Val 216 via hydrogen bonds and with Ala 60, Pro 98, Leu 99B, Phe 192, Ser 214, and Phe 215 via hydrophobic interactions (Figure 3). Pedunculagin was predicted to interact with leukocyte elastase via Asn 61, Val 216 via hydrogen bond and with Phe 41, Cys 42, His 57, Cys 58, Ala 60, Val 62, Leu 99B, Phe 192, Ser 214, and Phe 215 via hydrophobic interactions (Figure 4).

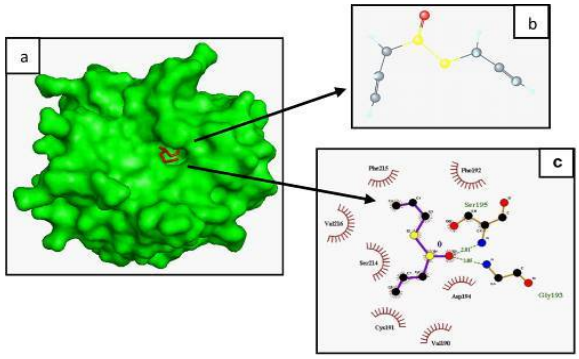

Figure 2: Molecular docking between target protein (leukocyte elastase) and allicin: (a) allicin (red) binds leukocyte elastase (green), (b) chemical structure of allicin, (c) interaction between allicin and target protein

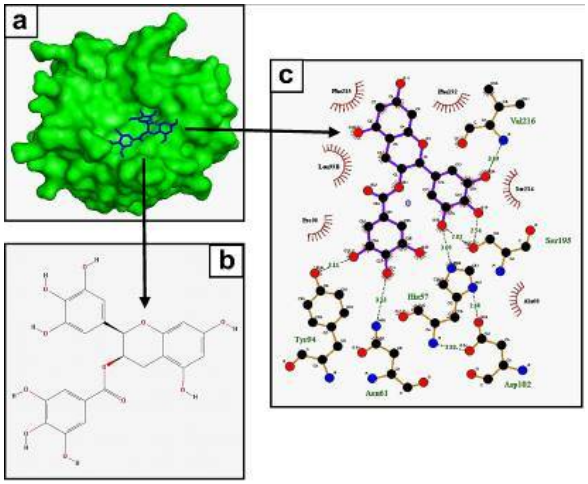

Figure 3: Molecular docking between target protein (leukocyte elastase) and epigalocathecin gallate. (a) epigalocathecin gallate (blue) bind to leukocyte elastase (green). (b) chemical structure of epigalocathecin gallate. (c) interaction between epigalocathecin gallate and target protein

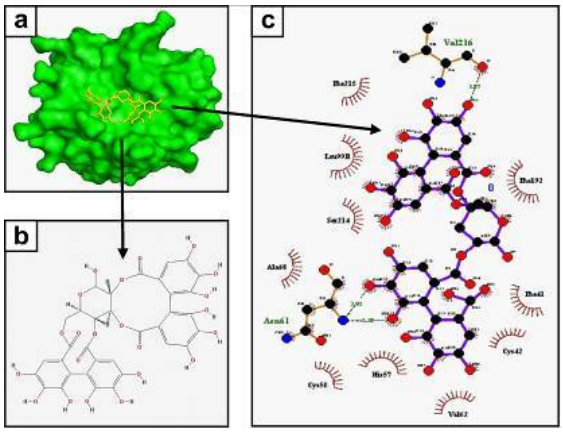

Figure 4: Molecular docking between target protein (leukocyte elastase) and pedunculagin. (a) pedunculagin (yellow) bind to leukocyte elastase (green). (b) chemical structure of pedunculagin. (c) interaction between pedunculagin and target protein

Furthermore, using reverse docking it was discovered that allicin could bind to leukocyte elastase at the same binding site as the known inhibitors of leukocyte elastase, epigallocatechin gallate and pedunculagin (Figure 5). Then, using DruLito software, we found that allicin fulfills all the Lipinski's rule parameters. So, allicin is a potential anti-aging drug according to Lipinski's rule parameter (Table 1).

DISCUSSION

Leukocyte elastase is one of the proteins that has role in the aging mechanism, by degrading extracellular matrix components such as elastin. The inhibition of leukocyte elastase activity can

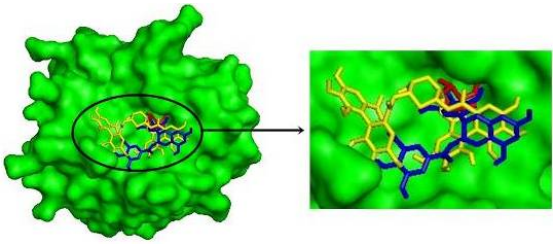

Figure 5: Interaction between leukocyte elastase and inhibitors (allicin, epigalocathecin gallate, and pedunculagin) show that inhibitors bind with leukocyte elastase at the same site. ***Key:*** green (leukocyte elastase), red (allicin), blue (epigalocathecin gallate), and yellow (pedunculagin)

Table 1: Lipinski's rule parameter to check compound's drug-likeness

Lipinski's rule / Compound	Molecular weight (<500 g/mol)	Log P (<5)	H-bond donor (<5)	H-bond acceptor (<10)
Allicin	162.02	0.237	0	1
Epigalocathecin gallate	458.08	2.984	8	11
Pedunculagin	759.89	-1.789	0	10

interrupt the apoptosis cycle and oxidative stress [21]. Inhibition of leukocyte elastase has experimentally shown to be a treatment for premature aging [22]. Skin diseases such as neurodermatitis, acne, and keratosis could be reduced by leukocyte activity [23]. Premature aging processes induces loss of skin elasticity due to extracellular matrix. Selective and effective inhibition of leukocyte elastase can be a promising strategy for drug development for the treatment of aging [24].

Reverse docking is a powerful tool for drug repositioning and drug discovery. It involves docking a small-molecule drug/ligand in the potential binding cavities of a set of clinically relevant macromolecular targets [25,26]. Binding affinity is an important aspect that must be considered when studying interactions between molecules and macromolecules [27]. A lower binding affinity signifies that the compound requires very little energy to do the binding or interaction. The value of a lower binding affinity increases the potential for binding to the target protein [28,29]. Reverse docking results showed that allicin is a very good inhibitor for leukocyte elatase [23].

Residues that have the interaction with leukocyte elastase include Phe 192, ser 214, Phe 215, and Val 216, which is part of the primary structure of leukocyte elastase [30]. Residues Phe 192 and Val 216 are an active site on the leukocyte

elastase, so it can be said that allicin is capable of binding and inhibiting the activity of leukocyte elastase [31].

Drug-like compounds must contain enough functionality to interact in a meaningful way with a protein [32], and the physicochemical properties of the compound will determine the quality of these interactions [33]. The physicochemical parameter ranges of Lipinski's rule-of-five is molecular weight < 500, log P < 5, H-bond donors < 5, H-bond acceptors < 10 [33-35].

CONCLUSION

The findings of this study indicate that allicin has the capacity to serve as a leukocyte elastase inhibitor based on its binding affinity and intermolecular interactions. Thus, allicin is a potential anti-aging drug candidate based on Lipinski's rule.

DECLARATIONS

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Conflict of interest

No conflict of interest is associated with this work.

Contribution of authors

The authors declare that this work was done by the authors named in this article. Ardini Pangastuti conducted the research and write the paper. Sri Endah Indriwati and Mohamad Amin designed the research and revise the paper. All authors read and approved the manuscript for publication.

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