

## IN SILICO ASSESSMENT OF AMELIORATIVE EFFECTS OF POLYUNSATURATED FATTY ACID (PUFAS) FROM *NAVICULA SALINICOLA* AS AN INHIBITOR OF BENIGN PROSTATE HYPERPLASIA

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### ABSTRACT

**Objective:** Benign prostatic hyperplasia (BPH) is a prevalent, non-cancerous condition affecting aging men worldwide. As an alternative approach to conventional treatment options, polyunsaturated fatty acids (PUFAs) have gained attention for their potential therapeutic effects on various health conditions. This study investigated the interaction of PUFAs obtained from *Navicula salinicola* with the macromolecule associated with BPH, represented by STAT3, that is involved in the androgen signaling pathway in BPH (PDB ID 6NJS), using molecular docking simulations.

**Methods:** The docking simulations revealed the interaction patterns and binding affinities of 14 PUFAs with the amino acid residues of STAT3. The calculated binding energies and inhibition constants provided insights into the potential inhibitory effects of PUFAs on BPH.

**Results:** Results indicated that  $\gamma$ -linolenic acid exhibited a strong binding affinity, forming hydrogen bonds with ARG609 and hydrophobic interactions with VAL637 and PRO639, highlighting its potential as a potent inhibitor. Docosahexaenoic acid also showed favorable interactions with ARG609 and hydrophobic residues, suggesting its potential therapeutic relevance.

**Conclusion:**  $\gamma$ -Linolenic acid from *N. salinicola* exhibited a strong molecular interaction with STAT3.

**Keywords:** Benign prostatic hyperplasia (BPH), Inhibitor, Molecular docking, *Navicula salinicola*, Polyunsaturated fatty acids (PUFAs)

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### INTRODUCTION

Benign prostatic hyperplasia (BPH) is a prevalent, non-cancerous condition that affects a significant number of aging men worldwide [1]. BPH involves the enlargement of the prostate gland, leading to the compression of the urethra and subsequent urinary symptoms such as difficulty in urination, increased frequency, and nocturia [2]. Despite being a non-life-threatening condition, BPH can significantly impact the quality of life and well-being of affected individuals. Currently, available treatment options for BPH include medications, minimally invasive procedures, and surgery [3]. However, minimally invasive surgery is still surgery, and there are risks of complications such as bleeding, infection, and injury to organs [4, 5]. Therefore, there is a growing interest in exploring alternative and potentially more effective therapeutic approaches.

The signal transducer and activator of transcription 3 (STAT3) is a protein that plays a role in the development of BPH. STAT3 is involved in the activation of NF- $\kappa$ B in the prostate as a result of high-fat diet feeding, leading to inflammation [6]. Constitutive activation of STAT3 causes growth dysregulation and resistance to apoptosis through changes in gene expression, leading to hyperproliferation of prostate tissue [7]. STAT3 is involved in the androgen signaling pathway in BPH [8]. The aberrant activation of STAT3 leads to increased cell proliferation and reduced cell apoptosis, resulting in BPH development [9]. The EGF/STAT3 signaling pathway is involved in the development of BPH, and qianliening capsule (QC) treatment may significantly decrease the serum level of EGF in BPH rats as well as downregulate the mRNA and protein levels in STAT3 [10].

Among the numerous natural compounds investigated for their potential health benefits, PUFAs have garnered attention due to their diverse physiological roles and potential therapeutic effects [11]. PUFAs, particularly omega-3 and omega-6 fatty acids, are essential nutrients obtained through diet and have been extensively studied for their cardiovascular, anti-inflammatory, and anticancer properties [12]. Researchers have also begun to investigate their potential impact on prostate health, particularly in the context of BPH [13].

In recent years, a specific microalgae called *N. salinicola* has emerged as a promising source of bioactive compounds, including various

PUFAs [14]. This microorganism thrives in saline environments and has been found to contain a rich reservoir of valuable lipids with potential health benefits [15]. *N. salinicola* is a species of diatom, a type of unicellular algae. Diatoms are known to be a potent source of PUFAs, which are essential for good health in humans and many animals [16]. *N. salinicola* has been studied for its fatty acid profile, and its potential as a source of PUFAs has been investigated [17]. Researchers have turned their attention to *N. salinicola* and its PUFA content to explore their therapeutic potential in various health conditions, including BPH.

The PUFAs found in *N. salinicola* have been of particular interest due to their potential as inhibitors of benign prostate hyperplasia [18]. Benign prostate hyperplasia is a non-cancerous enlargement of the prostate gland that can cause urinary problems in men. PUFAs have been studied for their potential to inhibit the growth of prostate cells and reduce the risk of benign prostate hyperplasia [19]. Most data regarding the effects of high dietary n-6 PUFA are positively associated with prostate cancer incidence. However, the ratio of n-6 to n-3 fatty acids may be more important than the absolute amount of n-3 PUFA [20]. PUFAs are associated with a reduced risk of several types of carcinogenesis, including prostate cancer. However, this depends on numerous factors, including the source of omega-3 PUFAs. The consumption of a high-fat diet and obesity cause a reduction in testosterone and promote prostatic changes such as prostatitis, BPH, and high-grade prostatic intraepithelial neoplasia (HGPIN) until cancer [21].

Despite these exciting developments, there remains a significant research gap about the molecular mechanisms underlying the potential inhibitory effects of PUFAs from *N. salinicola* on BPH [17]. Omega-3 PUFAs have been studied for their potential role in the treatment of benign prostatic hyperplasia (BPH). A study evaluated the effects of concentrated long-chain omega-3 polyunsaturated fatty acid (LCn3) supplementation on prostate cancer-specific quality of life in men treated by radical prostatectomy. The study found that LCn3 supplementation improved urinary incontinence and bowel function [22]. Another study investigated the effects of combination therapy with omega-3 fatty acids, tamsulosin, and finasteride in the treatment of men with lower urinary tract symptoms (LUTS) and BPH. The study found that the combination

therapy improved LUTS and BPH symptoms [23]. Omega-3 PUFAs have been shown to reduce blood pressure and vasodilation, which may be beneficial for men with BPH [24]. Supplementation of PUFAs and aerobic exercise have been shown to improve functioning, morphology, and redox balance in prostate-obese rats [21]. A study showed significantly lower omega-3 levels in men without prostate cancer, suggesting that omega-3 PUFAs may play a role in prostate cancer prevention [25]. While there have been some preliminary studies indicating the possible benefits of PUFAs in managing BPH, the exact molecular pathways and interactions between these bioactive compounds and prostate cells remain largely unexplored. Therefore, the present study aims to address this research gap through molecular docking analyses to gain deeper insights into the potential interactions between PUFAs from *N. salinicola* and the key molecules involved in BPH development.

Molecular docking is a computational technique that can be used to predict the binding affinity of a ligand to a receptor. In the field of BPH, molecular docking has been utilized in various research designs to investigate the potential of therapeutic targets and inhibitors for the treatment of BPH. Molecular docking can be used to identify potential therapeutic targets for the treatment of BPH. "A functional genomics pilot study" utilized molecular docking to identify alternative Chinese herbal agents for the treatment of BPH [26]. Molecular docking can be employed to investigate the binding interactions between ligands and receptors in BPH [2]. Molecular docking can be used to identify potential inhibitors for the treatment of BPH [27].

Molecular docking simulations were used to study the interactions between PUFAs from *N. salinicola* and the selected target molecules involved in BPH pathogenesis. These simulations will predict and analyze the binding affinities and binding modes of PUFAs with the target molecules, providing insights into potential inhibitory effects.

## MATERIALS AND METHODS

### Materials

The personal computer was equipped with an Intel® Core™ i7-7200U CPU at 2.50 GHz (4 CPUs at 2.7 GHz), 20 GB of RAM, and Windows 10 Pro 64-bit for molecular docking simulation.

### Preparation of ligand structures

The chemical structures of PUFAs (table 1), in a \*. sdf format file were obtained from the PubChem substance and compound databases (<https://pubchem.ncbi.nlm.nih.gov/>) [28]. The optimized 3D structures were converted using Discovery Studio Visualizer into \*. pdb file formats for further use in molecular docking simulations.

### Preparation of macromolecular structures

The crystal structure of the signal transducer and activator of transcription 3 (STAT3) (PDB ID 6NJS) along with the native ligand of SD36 CC(=O)N(C1=CC=C(C=C1)N(C2=CC=C(C=C2)N(C3=CC=C(C=C3)N(C4=CC=C(C=C4)N(C5=CC=C(C=C5)N(C6=CC=C(C=C6)N(C7=CC=C(C=C7)N(C8=CC=C(C=C8)N(C9=CC=C(C=C9)N(C10=CC=C(C=C10)N(C11=CC=C(C=C11)N(C12=CC=C(C=C12)N(C13=CC=C(C=C13)N(C14=CC=C(C=C14)N(C15=CC=C(C=C15)N(C16=CC=C(C=C16)N(C17=CC=C(C=C17)N(C18=CC=C(C=C18)N(C19=CC=C(C=C19)N(C20=CC=C(C=C20)N(C21=CC=C(C=C21)N(C22=CC=C(C=C22)N(C23=CC=C(C=C23)N(C24=CC=C(C=C24)N(C25=CC=C(C=C25)N(C26=CC=C(C=C26)N(C27=CC=C(C=C27)N(C28=CC=C(C=C28)N(C29=CC=C(C=C29)N(C30=CC=C(C=C30)N(C31=CC=C(C=C31)N(C32=CC=C(C=C32)N(C33=CC=C(C=C33)N(C34=CC=C(C=C34)N(C35=CC=C(C=C35)N(C36=CC=C(C=C36)N(C37=CC=C(C=C37)N(C38=CC=C(C=C38)N(C39=CC=C(C=C39)N(C40=CC=C(C=C40)N(C41=CC=C(C=C41)N(C42=CC=C(C=C42)N(C43=CC=C(C=C43)N(C44=CC=C(C=C44)N(C45=CC=C(C=C45)N(C46=CC=C(C=C46)N(C47=CC=C(C=C47)N(C48=CC=C(C=C48)N(C49=CC=C(C=C49)N(C50=CC=C(C=C50)N(C51=CC=C(C=C51)N(C52=CC=C(C=C52)N(C53=CC=C(C=C53)N(C54=CC=C(C=C54)N(C55=CC=C(C=C55)N(C56=CC=C(C=C56)N(C57=CC=C(C=C57)N(C58=CC=C(C=C58)N(C59=CC=C(C=C59)N(C60=CC=C(C=C60)N(C61=CC=C(C=C61)N(C62=CC=C(C=C62)N(C63=CC=C(C=C63)N(C64=CC=C(C=C64)N(C65=CC=C(C=C65)N(C66=CC=C(C=C66)N(C67=CC=C(C=C67)N(C68=CC=C(C=C68)N(C69=CC=C(C=C69)N(C70=CC=C(C=C70)N(C71=CC=C(C=C71)N(C72=CC=C(C=C72)N(C73=CC=C(C=C73)N(C74=CC=C(C=C74)N(C75=CC=C(C=C75)N(C76=CC=C(C=C76)N(C77=CC=C(C=C77)N(C78=CC=C(C=C78)N(C79=CC=C(C=C79)N(C80=CC=C(C=C80)N(C81=CC=C(C=C81)N(C82=CC=C(C=C82)N(C83=CC=C(C=C83)N(C84=CC=C(C=C84)N(C85=CC=C(C=C85)N(C86=CC=C(C=C86)N(C87=CC=C(C=C87)N(C88=CC=C(C=C88)N(C89=CC=C(C=C89)N(C90=CC=C(C=C90)N(C91=CC=C(C=C91)N(C92=CC=C(C=C92)N(C93=CC=C(C=C93)N(C94=CC=C(C=C94)N(C95=CC=C(C=C95)N(C96=CC=C(C=C96)N(C97=CC=C(C=C97)N(C98=CC=C(C=C98)N(C99=CC=C(C=C99)N(C100=CC=C(C=C100)N(C101=CC=C(C=C101)N(C102=CC=C(C=C102)N(C103=CC=C(C=C103)N(C104=CC=C(C=C104)N(C105=CC=C(C=C105)N(C106=CC=C(C=C106)N(C107=CC=C(C=C107)N(C108=CC=C(C=C108)N(C109=CC=C(C=C109)N(C110=CC=C(C=C110)N(C111=CC=C(C=C111)N(C112=CC=C(C=C112)N(C113=CC=C(C=C113)N(C114=CC=C(C=C114)N(C115=CC=C(C=C115)N(C116=CC=C(C=C116)N(C117=CC=C(C=C117)N(C118=CC=C(C=C118)N(C119=CC=C(C=C119)N(C120=CC=C(C=C120)N(C121=CC=C(C=C121)N(C122=CC=C(C=C122)N(C123=CC=C(C=C123)N(C124=CC=C(C=C124)N(C125=CC=C(C=C125)N(C126=CC=C(C=C126)N(C127=CC=C(C=C127)N(C128=CC=C(C=C128)N(C129=CC=C(C=C129)N(C130=CC=C(C=C130)N(C131=CC=C(C=C131)N(C132=CC=C(C=C132)N(C133=CC=C(C=C133)N(C134=CC=C(C=C134)N(C135=CC=C(C=C135)N(C136=CC=C(C=C136)N(C137=CC=C(C=C137)N(C138=CC=C(C=C138)N(C139=CC=C(C=C139)N(C140=CC=C(C=C140)N(C141=CC=C(C=C141)N(C142=CC=C(C=C142)N(C143=CC=C(C=C143)N(C144=CC=C(C=C144)N(C145=CC=C(C=C145)N(C146=CC=C(C=C146)N(C147=CC=C(C=C147)N(C148=CC=C(C=C148)N(C149=CC=C(C=C149)N(C150=CC=C(C=C150)N(C151=CC=C(C=C151)N(C152=CC=C(C=C152)N(C153=CC=C(C=C153)N(C154=CC=C(C=C154)N(C155=CC=C(C=C155)N(C156=CC=C(C=C156)N(C157=CC=C(C=C157)N(C158=CC=C(C=C158)N(C159=CC=C(C=C159)N(C160=CC=C(C=C160)N(C161=CC=C(C=C161)N(C162=CC=C(C=C162)N(C163=CC=C(C=C163)N(C164=CC=C(C=C164)N(C165=CC=C(C=C165)N(C166=CC=C(C=C166)N(C167=CC=C(C=C167)N(C168=CC=C(C=C168)N(C169=CC=C(C=C169)N(C170=CC=C(C=C170)N(C171=CC=C(C=C171)N(C172=CC=C(C=C172)N(C173=CC=C(C=C173)N(C174=CC=C(C=C174)N(C175=CC=C(C=C175)N(C176=CC=C(C=C176)N(C177=CC=C(C=C177)N(C178=CC=C(C=C178)N(C179=CC=C(C=C179)N(C180=CC=C(C=C180)N(C181=CC=C(C=C181)N(C182=CC=C(C=C182)N(C183=CC=C(C=C183)N(C184=CC=C(C=C184)N(C185=CC=C(C=C185)N(C186=CC=C(C=C186)N(C187=CC=C(C=C187)N(C188=CC=C(C=C188)N(C189=CC=C(C=C189)N(C190=CC=C(C=C190)N(C191=CC=C(C=C191)N(C192=CC=C(C=C192)N(C193=CC=C(C=C193)N(C194=CC=C(C=C194)N(C195=CC=C(C=C195)N(C196=CC=C(C=C196)N(C197=CC=C(C=C197)N(C198=CC=C(C=C198)N(C199=CC=C(C=C199)N(C200=CC=C(C=C200)N(C201=CC=C(C=C201)N(C202=CC=C(C=C202)N(C203=CC=C(C=C203)N(C204=CC=C(C=C204)N(C205=CC=C(C=C205)N(C206=CC=C(C=C206)N(C207=CC=C(C=C207)N(C208=CC=C(C=C208)N(C209=CC=C(C=C209)N(C210=CC=C(C=C210)N(C211=CC=C(C=C211)N(C212=CC=C(C=C212)N(C213=CC=C(C=C213)N(C214=CC=C(C=C214)N(C215=CC=C(C=C215)N(C216=CC=C(C=C216)N(C217=CC=C(C=C217)N(C218=CC=C(C=C218)N(C219=CC=C(C=C219)N(C220=CC=C(C=C220)N(C221=CC=C(C=C221)N(C222=CC=C(C=C222)N(C223=CC=C(C=C223)N(C224=CC=C(C=C224)N(C225=CC=C(C=C225)N(C226=CC=C(C=C226)N(C227=CC=C(C=C227)N(C228=CC=C(C=C228)N(C229=CC=C(C=C229)N(C230=CC=C(C=C230)N(C231=CC=C(C=C231)N(C232=CC=C(C=C232)N(C233=CC=C(C=C233)N(C234=CC=C(C=C234)N(C235=CC=C(C=C235)N(C236=CC=C(C=C236)N(C237=CC=C(C=C237)N(C238=CC=C(C=C238)N(C239=CC=C(C=C239)N(C240=CC=C(C=C240)N(C241=CC=C(C=C241)N(C242=CC=C(C=C242)N(C243=CC=C(C=C243)N(C244=CC=C(C=C244)N(C245=CC=C(C=C245)N(C246=CC=C(C=C246)N(C247=CC=C(C=C247)N(C248=CC=C(C=C248)N(C249=CC=C(C=C249)N(C250=CC=C(C=C250)N(C251=CC=C(C=C251)N(C252=CC=C(C=C252)N(C253=CC=C(C=C253)N(C254=CC=C(C=C254)N(C255=CC=C(C=C255)N(C256=CC=C(C=C256)N(C257=CC=C(C=C257)N(C258=CC=C(C=C258)N(C259=CC=C(C=C259)N(C260=CC=C(C=C260)N(C261=CC=C(C=C261)N(C262=CC=C(C=C262)N(C263=CC=C(C=C263)N(C264=CC=C(C=C264)N(C265=CC=C(C=C265)N(C266=CC=C(C=C266)N(C267=CC=C(C=C267)N(C268=CC=C(C=C268)N(C269=CC=C(C=C269)N(C270=CC=C(C=C270)N(C271=CC=C(C=C271)N(C272=CC=C(C=C272)N(C273=CC=C(C=C273)N(C274=CC=C(C=C274)N(C2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C(C=C801)N(C802=CC=C(C=C802)N(C803=CC=C(C=C803)N(C804=CC=C(C=C804)N(C805=CC=C(C=C805)N(C806=CC=C(C=C806)N(C807=CC=C(C=C807)N(C808=CC=C(C=C808)N(C809=CC=C(C=C809)N(C810=CC=C(C=C810)N(C811=CC=C(C=C811)N(C812=CC=C(C=C812)N(C813=CC=C(C=C813)N(C814=CC=C(C=C814)N(C815=CC=C(C=C815)N(C816=CC=C(C=C816)N(C817=CC=C(C=C817)N(C818=CC=C(C=C818)N(C819=CC=C(C=C819)N(C820=CC=C(C=C820)N(C821=CC=C(C=C821)N(C822=CC=C(C=C822)N(C823=CC=C(C=C823)N(C824=CC=C(C=C8

within an acceptable range of 2.0 Å, considering the inherent flexibility of both the ligand and the receptor during docking simulations. The RMSD value confirms the reliability of the docking methodology used in this study.

The molecular interactions between SD-36 and the macromolecule of signal transducer and activator of transcription 3 (STAT3) (PDB ID 6NJS) were successfully validated using Autodock 4.2. The RMSD value indicated that the docking simulations generated ligand poses in close agreement with the reference structure. The carefully chosen grid parameters and atom types ensured an accurate representation of the binding site and intermolecular interactions. This validation step supports the reliability and robustness of the docking methodology used in this study and provides a strong foundation for the subsequent analysis and interpretation of the docking results. The amino acid residues Glu-638 and Tyr-657 in the STAT3 SH2 domain have an important role in the activity of STAT3. These residues are involved in the binding of STAT3 to its target DNA and are potential targets for small-molecule inhibitors of STAT3 activity [35].

### Binding energies

We further investigated the binding interaction energies of various PUFAs with the macromolecule associated with benign prostatic

hyperplasia (BPH), represented by the PDB ID 6NJS. The interaction energies were calculated using Autodock 4.2, and the obtained binding energies and inhibition constants (Ki) for each PUFA are presented in table 1.

The binding energies represent the strength of the interactions between each PUFA and the active site of the macromolecule (6NJS). A higher binding energy indicates stronger and more favorable interactions, suggesting a higher likelihood of the PUFA forming stable complexes with the macromolecule [36]. Based on the binding energies, it can be seen that the native ligand (SD36) has the most favorable binding energy of -6.93 kcal/mol, indicating a strong and stable interaction with the macromolecule. This native ligand likely represents a biologically relevant compound involved in BPH regulation.

Among the PUFAs tested,  $\gamma$ -linolenic acid (-4.7 kcal/mol) and docosahexaenoic acid (-4.51 kcal/mol) also exhibit relatively high negative binding energies, suggesting that they have a strong affinity for the active site of 6NJS. This finding highlights their potential as potent inhibitors of BPH. On the other hand, several PUFAs, such as erucic acid, eicosatrienoic acid, and nervonic acid, show less favorable binding energies, indicating weaker interactions with the macromolecule.

**Table 1: Interaction energies of polyunsaturated fatty acids (PUFAs) in benign prostatic hyperplasia (PDB ID 6NJS)**

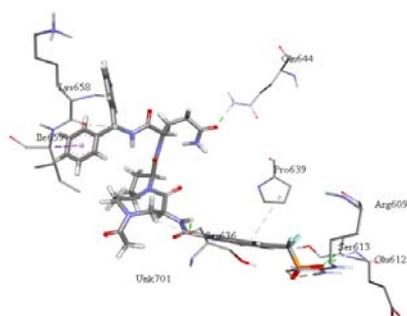
Code	Polyunsaturated fatty acids (PUFAs)	Binding energy, kcal/mol	Inhibition constant (Ki), mmol
SD36	Native ligand	-6.93	0.0083
ALA	$\alpha$ -Linoleic acid	-3.88	1.4400
ARA	Arachidonic acid	-3.94	1.3000
CHA	Cis-10-Heptadecenoic acid	-3.38	3.3200
DHA	Docosahexaenoic acid	-4.51	497.8400
EIA	Eicosatrienoic acid	-2.49	10.4500
ELA	Elaidic acid	-3.25	4.1400
EPA	Eicosapentaenoic acid	-3.16	4.8100
ERA	Erucic acid	-2.34	19.2300
GLA	$\gamma$ -Linolenic acid	-4.70	0.3575
HDA	Hexadecadienoic acid	-3.54	2.5500
MYA	Myristoleic acid	-3.64	2.1600
NEA	Nervonic acid	-2.75	9.5900
OLA	Oleic acid	-3.14	4.9800
PAA	Palmitoleic acid	-3.59	2.3200

The inhibition constant (Ki) represents the concentration of the PUFA required to achieve 50% inhibition of the macromolecule's activity. A lower Ki value indicates a more potent inhibitor. The study found that  $\gamma$ -linolenic acid was a potent inhibitor with a Ki of 0.35746 mmol, followed by the native ligand (SD36) with a Ki of 0.00833 mmol.

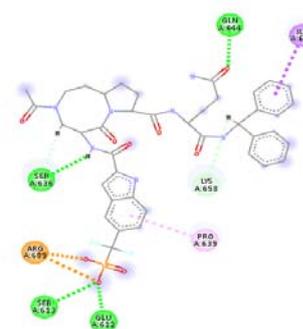
It's worth noting that while some PUFAs show favorable binding energies and low Ki values, others exhibit weaker interactions. These results suggest that not all PUFAs may have significant inhibitory effects on BPH, and their potential as therapeutic agents should be further investigated in experimental settings.

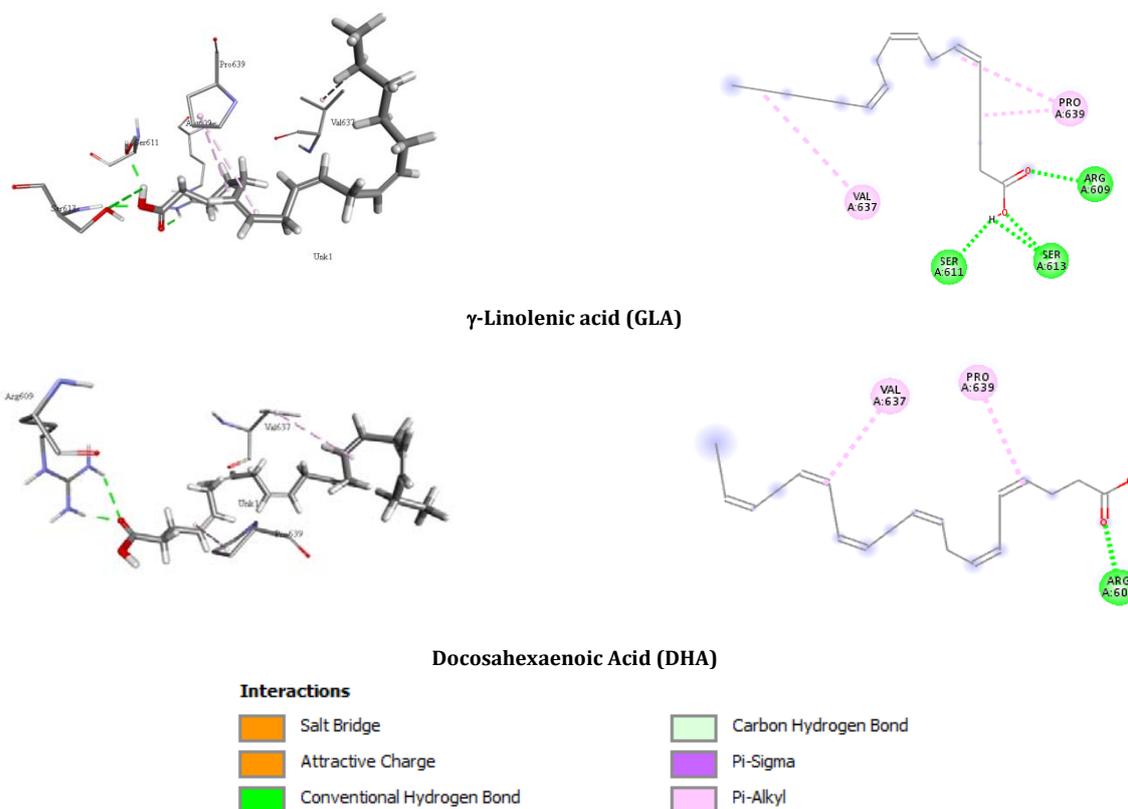
### Binding mode

Interaction analysis was performed to investigate the binding patterns between the top PUFAs and the amino acid residues of the macromolecule associated with BPH, represented by the PDB ID 6NJS. Analysis was conducted using Discovery Studio Visualizer, and the obtained data on hydrogen bonds (HB) and hydrophobic interactions (HI) between PUFAs and amino acid residues are presented in fig. 2. The interaction analysis provides valuable insights into the key residues involved in the binding of PUFAs to the active site of the macromolecule (6NJS), shedding light on the potential binding modes and stabilization of the complexes.



**Native ligand (SD36)**





**Fig. 2: A 2D and 3D view of the bond between top-two of the best-pose of PUFAs into amino acid residues of benign prostatic hyperplasia (PDB ID 6NJS)**

Hydrogen bonds are important non-covalent interactions that play a pivotal role in ligand-receptor binding. The native ligand (SD36) forms four hydrogen bonds with specific amino acid residues of the macromolecule, namely GLU612, SER613, SER636, and GLN644. These hydrogen bonds contribute significantly to the stability of the ligand-receptor complex. Besides,  $\gamma$ -linolenic acid and docosahexaenoic acid also form a hydrogen bond pattern similar to SD-36. Tetapi,  $\gamma$ -linolenic acid, and docosahexaenoic acid also form hydrogen bonds with the same amino acid residue, ARG609. This indicated that interaction suggests a common binding mode for these PUFAs within the active site of the macromolecule.

Hydrophobic interactions involve *van der Waals* forces between non-polar regions of the ligand and the receptor.  $\gamma$ -Linolenic acid and docosahexaenoic acid engage in two hydrophobic interactions each, involving the amino acid residues VAL637 and PRO639. These hydrophobic interactions contribute to the stability of the complexes by minimizing unfavorable solvent exposure to hydrophobic ligands and receptor regions.

Overall, interaction analysis showed the importance of certain amino acid residues in stabilizing the binding of PUFAs to BPH-related macromolecules. Hydrogen bonding and hydrophobic interactions play an important role in mediating these interactions. The identification of key residues involved in the binding of PUFAs provides valuable information for future structure-based drug design and the development of potential therapeutic agents for BPH.

## DISCUSSION

*N. salinicola* has been studied for its fatty acid profile and its potential as a source of PUFAs [37]. The role of *N. salinicola* in the production of PUFAs is related to its ability to synthesize these compounds through specific pathways [38]. The fatty acid profile of *N. salinicola* is similar to that of other diatoms, with 14:0, 16:0, 16:1*n*-7, and 20:5*n*-3 (EPA), being the major fatty acids for polar and neutral lipids. The strong concentration of 16:1*n*-7 and its high

enrichment supported its central role as a precursor of the C16 PUFA pathway. The C16 PUFA pathway is initiated by 16:1*n*-7, which is first desaturated in 16:2*n*-7 or 16:2*n*-4.

The position of the double bond in the fatty acid chain affects PUFA binding energy. PUFAs with a greater number of double bonds tend to exhibit more negative (lower) binding energies. This means that they have stronger interactions with the target macromolecule. This was because double bonds can enhance the flexibility and conformational adaptability of fatty acids, thereby allowing them to better fit into the binding pockets of the macromolecule.

The position of the first double bond from the omega end (methyl end) of the fatty acid chain also plays a role. Omega-3 PUFAs (e. g.,  $\alpha$ -linoleic acid, docosahexaenoic acid, and eicosapentaenoic acid) often exhibit more negative binding energies compared to omega-6 PUFAs (e. g., arachidonic acid and  $\gamma$ -linolenic acid). Omega-3 PUFAs are often linked to anti-inflammatory effects. Their extended carbon chain and higher number of double bonds may contribute to stronger interactions with the target, resulting in lower binding energies [39].

Monounsaturated fatty acids with a single, double bond (e. g., oleic acid and palmitoleic acid) tend to have moderate binding energies. While they have one double bond, their overall flexibility might be slightly limited compared to PUFAs with multiple double bonds. Elaidic acid, a *trans*-fatty acid, also demonstrates moderate binding energy. The *trans* configuration of the double bond could influence the interaction with the target, resulting in relatively moderate binding energy. The *cis*-10-heptadecenoic acid, with one double bond, displays moderate binding energy. The specific position of the double bond and its influence on interactions with the target contribute to this energy level.

In summary, the position and number of double bonds in PUFAs significantly impact their binding energy. Even though the binding energy of the omega-3 PUFAs was less than that of the native ligand,

PUFAs have advantages. Omega-3 PUFAs are a type of polyunsaturated fatty acid that is known for its anti-inflammatory effects and can be obtained from plant-based and marine sources. They have been shown to reduce the risk of cardiovascular disease, improve cognitive function, and improve depression [12]. PUFAs with higher numbers of double bonds and omega-3 configurations often exhibit stronger interactions with the target, resulting in more negative binding energies. The relationship between fatty acid structure and binding energy underscores the importance of understanding molecular interactions for potential therapeutic applications.

## CONCLUSION

Molecular docking was used for *in silico* investigation of the inhibitory effect of benign prostate hyperplasia on STAT3 protein by 15 PUFAs from *N. salinicola*. PUFAs from *N. salinicola* were found to interact with the macromolecule STAT3 (PDB ID 6NJS).  $\alpha$ -Linolenic acid and docosahexaenoic acid had substantial binding affinities for certain amino acid residues in the macromolecule's active site. Notably,  $\alpha$ -linolenic acid exhibited strong hydrogen bonding and hydrophobic interactions, indicating its potential as a potent BPH inhibitor. These findings suggest that natural substances like PUFAs could be used as innovative and targeted treatment agents for BPH control. However, more research is needed to demonstrate the inhibitory effects of PUFAs on BPH and to understand the underlying molecular dynamics.

## FUNDING

Nil

## AUTHORS CONTRIBUTIONS

All the authors have equally contributed to the current study.

## CONFLICT OF INTERESTS

All the authors declare no conflicts of interest.

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