Title: Flaxseed (Linum usitatissimum) Modulates Oxytocin Signaling Pathway via CALM3-Mediated Calcium Signaling: A PPI Network and Functional Enrichment Analysis

Running Title: Flaxseed's Anxiolytic Action via Oxytocin

Authors: Hajar Sadat Sadeghi[1], Mostafa Rezaei Tavirani[2,]*, Melika Abrishami[2], Maryam Hamzeloo-Moghadam[1]

1- *Traditional Medicine and Materia Medica Research Center, Department of Traditional Pharmacy, School of Traditional Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran.*

2- *Proteomics Research Center, Faculty of Paramedical Sciences, Shahid Beheshti University of Medical Sciences, Tehran, Iran.*

***Corresponding Author**: Mostafa Rezaei Tavirani, Proteomics Research Center, Faculty of Paramedical Sciences, Shahid Beheshti University of Medical Sciences, Tehran, Iran. Email: Tavirany@yahoo.com

To appear in: **Basic and Clinical Neuroscience**

Received date: 2025/05/26

Revised date: 2025/10/15

Accepted date: 2025/10/19

Please cite this article as:

Sadeghi, H.S., Rezaei Tavirani, M., Abrishami, M., Hamzeloo-Moghadam, M. (In Press). Flaxseed (Linum usitatissimum) Modulates Oxytocin Signaling Pathway via CALM3-Mediated Calcium Signaling: A PPI Network and Functional Enrichment Analysis. Basic and Clinical Neuroscience. Just Accepted publication Jul. 10, 2025. Doi: http://dx.doi.org/10.32598/bcn.2025.8066.1

DOI: http://dx.doi.org/10.32598/bcn.2025.8066.1

Abstract

Purpose: This study aimed to elucidate the molecular mechanism underlying the anti-anxiety effects of flaxseed (*Linum usitatissimum*) by investigating its role in modulating the oxytocin signaling pathway through protein-protein interaction (PPI) network analysis.

Methods: Gene expression profiles from the GEO dataset (GSE36422) were analyzed to identify flaxseed-induced differentially expressed genes (DEGs). Bioinformatics tools, including functional enrichment (KEGG, GO) and PPI network construction (STRING, Cytoscape), were used to map key genes and pathways. Hub and bottleneck proteins were identified using Cyto-Hubba's Maximal Clique Centrality (MCC) and Betweenness algorithms, with a focus on genes shared between flaxseed and oxytocin signaling.

Results: Of 98 initial DEGs, 56 were significantly regulated by flaxseed. Three critical genes—CALM3 (upregulated), NFATC4, and RAF1 (both downregulated)—emerged as shared mediators between flaxseed and oxytocin pathways. CALM3, a calcium-sensing hub protein, exhibited extensive network connectivity, interacting with 58% of oxytocin pathway proteins and functioning as a bottleneck regulator. Pathway analysis revealed flaxseed's influence on oxytocin signaling, cellular senescence, and cGMP-PKG pathways. NFATC4 downregulation suggested disinhibition of oxytocin signaling, while RAF1's role in MAPK cascades further supported anxiolytic effects.

Conclusion: Flaxseed exerts its anti-anxiety effects primarily through CALM3-mediated oxytocin signaling, leveraging calcium-dependent pathways. These findings provide mechanistic evidence supporting flaxseed as a natural anxiolytic and highlight its potential as a dietary intervention for anxiety disorders.

Key words: *Linum usitatissimum*, oxytocin signaling, anxiety, CALM3, PPI network

Introduction

Linum usitatissimum (Linn.), commonly known as flax, belongs to the Linaceae family (Watson & Preedy, 2012). This species is recognized for its striking pale blue flowers, which consist of five distinct petals. The seeds produced by the flax plant, referred to as flaxseeds, are characterized by their flat, pointed, oval shape (Ansari et al., 2019). The entire plant is utilized commercially, either in its raw state or after undergoing processing (Singh et al., 2011). Flaxseed has been incorporated into the human diet since the dawn of the first civilizations. In ancient Egypt and Greece, it was used for medicinal purposes (Service et al., 1990). Flaxseeds are known for their significant nutritional benefits and are a major source of omega-3 fatty acids, particularly α-linolenic acid (ALA). They are also abundant in short-chain polyunsaturated fatty acids (PUFAs), both soluble and insoluble fibers, phytoestrogenic lignans such as secoisolariciresinol diglycoside (SDG), proteins, and a range of antioxidants (Ivanov et al., 2011; Oomah, 2001; Singh et al., 2011; Touré & Xueming, 2010). The properties of these compounds make *L. usitatissimum* a suitable choice for addressing various health concerns, including respiratory issues, neurological disorders, diabetes, gastrointestinal problems, constipation, abdominal pain, urinary infections, and skin inflammations (Al-Madhagy et al., 2023; Mueed et al., 2022). A notable feature of flaxseed, as demonstrated by multiple studies, is its capacity to reduce anxiety (Al-Madhagy et al., 2023; Anis et al., 2016).

Generalized anxiety disorder (GAD) is a persistent and incapacitating condition that impacts roughly 6.1% of people throughout their lives. Anxiety is defined by sensations of fear, unease, and discomfort. It may present physically with symptoms such as perspiration, restlessness, muscle tension, and an elevated heart rate (DeMartini et al., 2019). Available treatment options encompass psychological therapies and pharmacological interventions (Hoge et al., 2012). Global guidelines for the management of GAD recommend selective serotonin reuptake inhibitors (SSRIs), serotonin and noradrenaline reuptake inhibitors (SNRIs), and pregabalin as primary treatment options, due to their proven effectiveness and favorable safety profiles (Bandelow et al., 2023).

Oxytocin, a neuropeptide composed of nine amino acids, is recognized as the first human hormone to be identified (Carter, 2022). This molecule is particularly noted for its role in stimulating uterine contractions. Furthermore, oxytocin has been proposed as an anti-anxiety agent in various studies (Gully et al., 2024). While oxytocin is released under various physiological conditions, research has indicated that its secretion can also be triggered by external stimuli, such as flaxseed extract (Lecová et al., 2024; Sirotkin, 2023).

At present, natural products represent a substantial portion of modern pharmaceutical agents, especially in the context of disease treatments (Pal & Shukla, 2003). A unique strategy to deepen our comprehension of the therapeutic mechanisms of active substances entails forecasting the gene networks influenced by the active compounds present in medicinal plants (Shao & Zhang, 2013). The analysis of protein-protein interactions (PPI) is beneficial for pinpointing this essential aspect (Tang et al., 2023). PPI network analysis elucidates the connections among proteins within a biological framework (Rezaei-Tavirani et al., 2022). Although the literature has documented the anti-anxiety effects of flaxseed, its molecular mechanism is still not fully understood. This research aims to demonstrate its molecular mechanism through the analysis of protein-protein interaction networks.

Materials and Methods

This study examined the gene expression profiles triggered by flaxseed through a thorough bioinformatics methodology. Gene expression data were meticulously gathered from the Gene Expression Omnibus (GEO) dataset GSE36422, utilizing GEO2R for the comparative analysis of expressed genes among various experimental groups. The initial data processing included a detailed Venn diagram visualization to clarify the gene expression profile uniquely linked to flaxseed intervention (Fig. 1). Bioinformatics techniques were employed to clarify the molecular mechanisms that underlie gene expression induced by flaxseed.

A comprehensive analytical approach was adopted, beginning with functional enrichment analysis through KEGG pathway and gene ontology (GO) methodologies. To gain a thorough understanding of the molecular interactions, the genes that overlap between flaxseed differentially expressed genes (DEGs) and oxytocin signaling pathways were subsequently analyzed. To validate the targeted genes, a protein-protein interaction (PPI) network for the oxytocin signaling pathway was constructed using STRING (version 11.5), with interaction confidence carefully filtered to a minimum score of 0.4. Following this, network analysis was performed. Cytoscape (version 3.10.3) was utilized, employing CytoHubba's MCC algorithm to rank hub proteins, while Betweenness Centrality was used to identify bottlenecks (threshold: top 5 nodes), and functional analysis was conducted with the ClueGo plugin, utilizing a kappa score of 0.4. Furthermore, the key genes associated with oxytocin in the context of flaxseed were investigated within the network.

Results

The Venn diagram depicting gene expressions induced by flaxseed from GSE36422 illustrates a total of 98 significant gene expressions linked to flaxseed, with 56 genes retained after data cleaning. These 56 genes underwent further analysis. The data were assessed through Venn diagrams comparing Radiation–(Flaxseed-Radiation), Control-Radiation, Flaxseed-Control, and (Flaxseed-Radiation)-Flaxseed groups. The Venn diagram revealed that 98 significantly differentially expressed genes (DEGs) differentiate the flaxseed group from the control groups (Fig. 1). Analysis utilizing a volcano plot indicated a significant disparity in gene expression profiles between samples subjected to a 10% flaxseed diet for three weeks (Flaxseed group) and untreated samples. The box plot for the Flaxseed-Control comparison demonstrated that all gene expression profiles were median-centered and comparable. Likewise, the density plot exhibited a consistent pattern in the Flaxseed-Control analysis. After eliminating duplicate genes and uncharacterized entries, the 98 significant DEGs were narrowed down to 56, representing the genes significantly influenced by the presence of flaxseed.

Pathway analysis identifies three significant genes: CALM3 (Calmodulin3), NFATC4 (Nuclear Factor of Activated T Cells 4), and RAF1 (Raf-1 Proto-Oncogene), which are prevalent in both the flaxseed and oxytocin signaling pathways (refer to Fig. 2 and Fig. 3). KEGG pathway enrichment analysis reveals several pathways characterized by high counts and the most elevated logP values, including the C-type lectin receptor signaling pathway, the oxytocin signaling pathway, cellular senescence, and the cGMP-PKG signaling pathway. These genes are crucial across all these pathways, with CALM3 being up-regulated (overexpressed) while NFATC4 and RAF1 are down-regulated (underexpressed).

The gene ontology analysis, depicted with various colored lines, illustrates distinct biological pathways, where the size of each node reflects the relative significance or connectivity of each gene within the network. CALM3 is represented as a red node, indicating its involvement in multiple signaling pathways such as Cellular senescence, Oxytocin signaling, C-type lectin receptor signaling, and GnRH signaling, while also showing associations with viral infection pathways (specifically HIV-1 and cytomegalovirus). The color coding indicates that it is upregulated (represented in red on the log2 fold change scale). NFATC4, depicted as a purple node within the network, is linked to pathways associated with: Long-term potentiation, Viral infections, and GnRH signaling. This transcription factor is essential for immune response and cellular development. The color coding indicates that it is downregulated (purple in the log2 fold change scale). RAF1, illustrated as a blue node, is significantly connected to various pathways: cGMP-PKG signaling, responses to viral infections, and Long-term potentiation. The color coding also suggests it is downregulated (purple in the log2 fold change scale) (Fig. 2). The genes related to the oxytocin signaling pathway include CALM3, NFATC4, and RAF1, with a p-adj value of 0.0017. RAF1 is involved in the MAPK signaling pathway, CALM3 regulates calcium channels, and NFATC4 negatively regulates the OXT pathway (Fig. 2). The precise positioning of each gene within the Oxytocin signaling Pathway is as follows: RAF1 is integrated into the MAPK signaling pathway, activated downstream of Ras, which is stimulated by the activation of the oxytocin receptor (OXTR). It phosphorylates and activates MEK1/2, which subsequently activates ERK1/2. The activation of ERK1/2 results in the transcriptional regulation of genes that are crucial for Proliferation, Differentiation, and the survival of Cardiomyocytes. RAF1 is vital in enhancing the parturition cascade (the process of uterine contraction during labor). NFATC4 operates downstream of calcium signaling, activated by calcium-dependent signaling via calcineurin (CALN). Once dephosphorylated by calcineurin, NFATC4 migrates to the nucleus to influence gene expression. It plays a role in the negative regulation of oxytocin signaling in myometrial cells, regulating genes such as RCAN1 and RGS2, which modulate oxytocin receptor signaling and calcium dynamics. CALM3 plays a pivotal role in calcium signaling by binding to calcium ions, thereby functioning as a calcium sensor. Additionally, it activates downstream targets such as CaMK (calmodulin-dependent kinase) and Calcineurin (CALN). Through these interactions, CALM3 exerts regulatory control over MLCK (myosin light chain kinase), which is essential for muscle contraction in both myometrial and mammary gland cells. In summary, RAF1 is instrumental in mediating MAPK signaling pathways that facilitate proliferation and differentiation. NFATC4 is responsible for regulating transcription in response to calcium signaling, thereby modulating oxytocin signaling. CALM3 serves as a calcium sensor that governs various downstream pathways, including those involved in contraction and metabolism. These genes are vital for the physiological responses triggered by oxytocin, which encompass uterine contraction, cardiovascular effects, and cellular proliferation (Fig. 3). A collection of 147 proteins associated with the oxytocin signaling pathway was assembled utilizing STRING and subsequently examined in Cytoscape. Summary Statistics of the network: Number of nodes: 147, Number of edges: 2172, Average number of neighbors: 29.551, Network density: 0.202, Network heterogeneity: 0.644, Network centralization: 0.406. Among these proteins, 86 (highlighted in red) are the primary neighbors of CALM3, and in total, 91 out of the 147 proteins are the primary neighbors of the three specified genes. The top five hub and bottleneck proteins were determined using the Cyto-Hubba plugin (refer to Tables 1 and 2). Within the first neighbor nodes, there exists one hub protein (denoted by a diamond shape) and five bottleneck proteins, including CALM3 (denoted by rectangle shapes) (see Fig. 4).

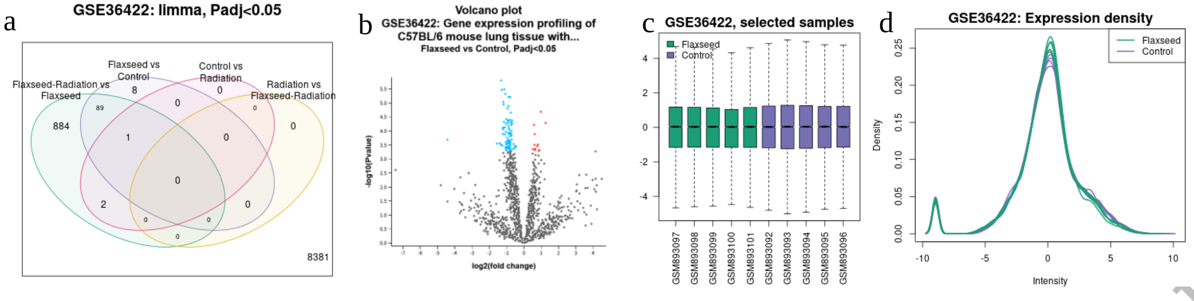

Fig. 1: The Flaxseed-induced gene expressions of GSE36422, a) Venn diagram for radiation – (Flaxseed - Radiation), Control - Radiation, Flaxseed - Control, and (Flaxseed - Radiation) - Flaxseed analyses. b) Volcano plot of gene expression analysis of the Flaxseed group versus samples without any treatment (Control group). c) Box plot of Flaxseed – Control analysis. d) Expression density for Flaxseed – Control analysis

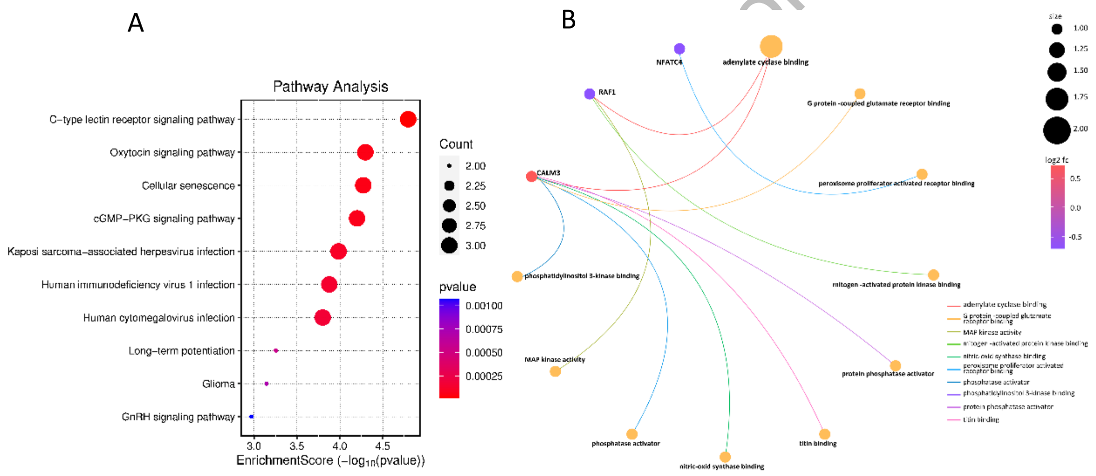

Fig. 2: Functional enrichment analysis of 56 selected genes expressions of flaxseed. A) KEGG pathway enrichment analysis for flaxseed genes. B) Gene Ontology (GO) enrichment analysis categorizes biological processes

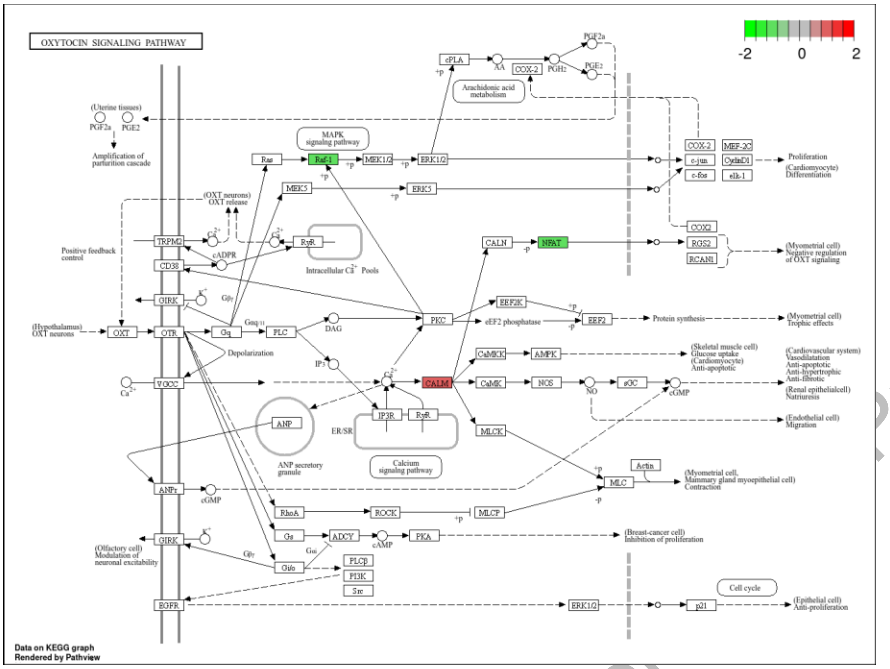

Fig. 3: The KEGG graph of the oxytocin signaling pathway

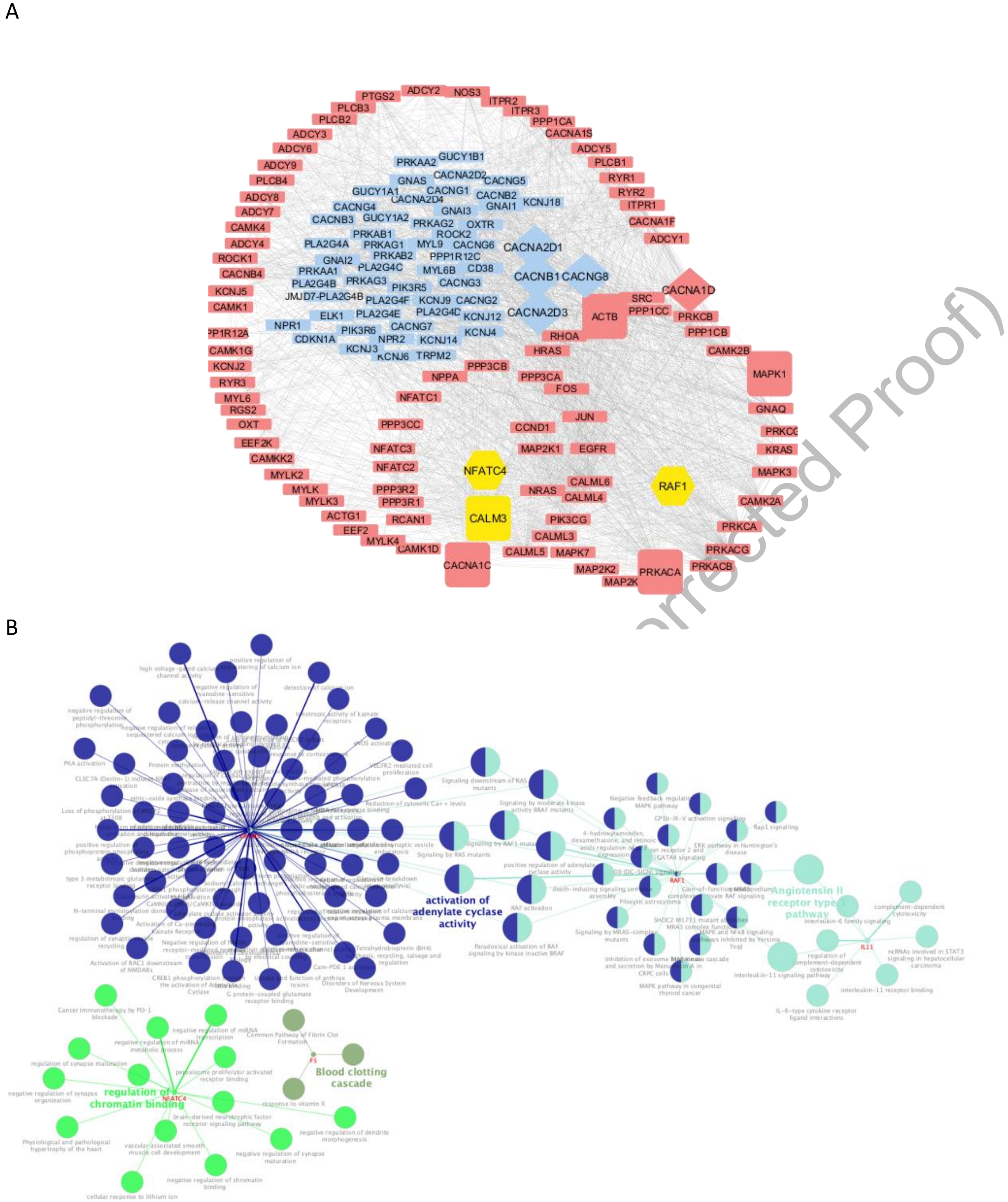

Fig. 4: A) The PPI network of the Oxytocin pathway, the yellow nodes are the common genes with flaxseed, and the red nodes are the first neighbors of CALM3, the hub nodes are in a diamond shape, and the bottlenecks are rectangular. B) This network illustrates the functional enrichment of biological pathways derived from genes modulated by flaxseed, constructed using the ClueGO plugin. Each node represents a significantly enriched pathway. Edges indicate gene overlap between pathways, highlighting potential mechanistic crosstalk. The green cluster, anchored by the core gene NFATC4, highlights pathways associated with the regulation

of chromatin binding, and synaptic signaling modulation. The dark blue cluster, dominated by CALM3, centers on activating adenylate cyclase activity, and calcium-mediated signal transduction. A light blue cluster, merging with dark blue elements and linked by RAF1, underscores the involvement of MAPK and RAS signaling cascades, pointing to flaxseed's potential influence on cell proliferation and survival mechanisms. The light cyan cluster, anchored by IL11, which as well reflects involvement in inflammatory and cytokine-mediated pathways, including the Interleukin-11 signaling pathway and Angiotensin II receptor type 1 pathway.

Table 1. The top 5 hub proteins ranked by the Maximal Clique Centrality (MCC) algorithm. Scores represent MCC values; higher scores indicate greater hubness

Rank	Name	Score	BetweennessCentrality	Degree
1	CACNA1D	5.40E+22	0.600823045	54
2	CACNA2D1	5.40E+22	0.530909091	33
3	CACNB1	5.40E+22	0.528985507	31
3	CACNG8	5.40E+22	0.525179856	30
3	CACNA2D3	5.40E+22	0.506944444	29

Table 2. The Top 5 bottleneck proteins ranked by Betweenness Centrality. Scores reflect Betweenness values; higher scores show higher bottleneck influence.

Rank	Name	Score	BetweennessCentrality	Degree
1	CALM3	36	0.701923077	85
2	MAPK1	19	0.591093117	55
3	CACNA1C	16	0.610878661	57
4	PRKACA	15	0.712195122	88
5	ACTB	10	0.577075099	45

Discussion

Anxiety is recognized as one of the most prevalent mental disorders within society. In the context of Iranian traditional medicine literature, anxiety is perceived in a distinct manner (Kaplan et al., 2007). The term "Khuf" refers to the unease concerning a potentially adverse situation that may occur in the future (Motovaseliyan et al., 2016). Avicenna identified flaxseed as a valuable therapeutic approach for managing anxiety (Avecinna, 2005). Flaxseed (*Linum usitatissimum* L., part of the Linaceae family), which is abundant in biologically active compounds, is acknowledged for its nutritional, physiological, and therapeutic properties (Sirotkin, 2023). Research conducted both in vivo and in vitro has provided evidence of hormonal regulation linked to flaxseed, including the stimulation of oxytocin expression (Sirotkin, 2023; Tou et al., 1999). In their study, Lecova et al. found that pigs fed a diet enriched with 10% flaxseed showed increased levels of oxytocin (Lecová et al., 2024). Additionally, research by Vlčková et al. indicated that flaxseed has a positive impact on the physiological effects of oxytocin by offering a protective mechanism for oxytocin receptors in the presence of xylene (Vlčková et al., 2022). In this investigation, we illustrated the regulation of various biological pathways by flaxseed-induced differentially expressed genes (DEGs), which include the C-type lectin receptor signaling pathway, the oxytocin signaling pathway, cellular senescence, and the cGMP-PKG signaling pathway, as determined by Functional Enrichment Analysis. The anti-anxiety properties of oxytocin are well-documented and extensively researched in medical sciences. Tang et al. demonstrated that the intranasal administration of oxytocin alleviated post-surgical anxiety in mice (Tang et al., 2024). Furthermore, a study by Chaipunko et al. revealed that oxytocin can influence the functioning of the hypothalamic-pituitary-adrenal axis (HPA axis) and mitigate anxiety (Chaipunko et al., 2025).

GO analysis identified three genes (CALM3, RAF1, NFACT4) among 56 flaxseed-induced DEGs as the most significant and relevant common genes linking flaxseed to the oxytocin pathway. Among these three pivotal genes, CALM3 stands out as the most crucial participant, exhibiting distinct characteristics and regulatory behaviors. In contrast to its counterparts, RAF1 and NFATC4, which displayed downregulation, CALM3's upregulation, along with its remarkable position within the network, designates it as a vital mediator in this molecular cascade. This interpretation is particularly reinforced by the established role of NFATC4 as a negative regulator of the oxytocin pathway. Consequently, its downregulation is anticipated to lead to diminished inhibition and, as a result, enhanced oxytocin signaling. The network analysis further highlighted CALM3 as a bottleneck, sustaining first-neighbor interactions with five other essential proteins (four bottleneck proteins and one hub protein). Furthermore, it showcases remarkable connectivity with 85 first neighbors, which accounts for 58% of all proteins involved in the oxytocin signaling pathway. This approach implies that CALM3 functions as a master regulator within the network, orchestrating various signaling cascades and ensuring the proper flow of information throughout the system. The significant impact of CALM3 within the network strongly supports its position as the primary molecular mediator, through which flaxseed may exert its anxiolytic effects via the oxytocin pathway. Oxytocin, a neuropeptide that plays a role in social bonding, reproductive functions, and stress responses, primarily exerts its effects by activating its receptors, resulting in an increase in intracellular calcium levels. The rise in calcium activates CALM3, which facilitates downstream signaling processes such as the activation of the MAPK pathway and the RhoA/Rho kinase pathway, both of which are associated with smooth muscle contraction and other physiological responses to oxytocin (Bakos et al., 2018; Munk et al., 2022; Uvnaes Moberg et al., 2022). Moreover, CALM3 may also affect cardiovascular functions regulated by oxytocin, underscoring its potential role in integrating calcium signaling with the effects of oxytocin on the cardiovascular system (Ayar et al., 2014; Hirasawa et al., 2001; Rabow et al., 2023). Therefore, CALM3 acts as a vital link between calcium signaling and the physiological effects of oxytocin. Additionally, it offers potentially significant insights for therapeutic strategies in anxiety management through dietary interventions.

Conclusion

This research clarifies a new mechanism through which flaxseed influences anxiety by way of the oxytocin pathway.

In silico analysis identifies the involvement of three common genes associated with both anxiety and flaxseed. These genes are crucial in the oxytocin signaling pathway, with CALM3 identified as a primary calcium-sensing regulator.

Simultaneously, the downregulation of NFATC4 and RAF1 further facilitates the disinhibition of oxytocin signaling and the modulation of the MAPK cascade. These results not only affirm the traditional applications of flaxseed but also underscore its potential as a dietary supplement for anxiety disorders. Future investigations should confirm these mechanisms through clinical trials and examine the synergistic effects alongside conventional treatments.

Acknowledgements

This article was derived from the PhD thesis on Traditional Pharmacy by Dr. Hajar Sadat Sadeghi, Shahid Beheshti University of Medical Sciences, Tehran, Iran, Grant No. 43006118.

References

Al-Madhagy, S., Ashmawy, N. S., Mamdouh, A., Eldahshan, O. A., & Farag, M. A. (2023). A comprehensive review of the health benefits of flaxseed oil in relation to its chemical composition and comparison with other omega-3-rich oils. *European Journal of Medical Research*, *28*(1), 240. https://doi.org/10.1186/s40001-023-01203-6

Anis, L., Haider, S., & Sajid, I. (2016). Repeated oral administration of flaxseeds induced antidepressant and anxiolytic effects in rats. *FUUAST Journal of Biology*, *6*(1), 41-46.

Ansari, R., Zarshenas, M. M., & Dadbakhsh, A. H. (2019). A Review on Pharmacological and Clinical Aspects of Linum usitatissimum L. *Current drug discovery technologies*, *16*(2), 148-158.

Avecinna. (2005). *Al-Qanoon fi-teb* (1 ed.). Iran university of medical sciences.

Ayar, A., Ozcan, M., Alcin, E., Serhatlioglu, I., Ozcan, S., Kutlu, S., & Kelestimur, H. (2014). Oxytocin activates calcium signaling in rat sensory neurons through a protein kinase C-dependent mechanism. *Journal of physiology and biochemistry*, *70*, 43-48.

Bakos, J., Srancikova, A., Havranek, T., & Bacova, Z. (2018). Molecular mechanisms of oxytocin signaling at the synaptic connection. *Neural plasticity*, *2018*(1), 4864107.

Bandelow, B., Allgulander, C., Baldwin, D. S., Costa, D. L. d. C., Denys, D., Dilbaz, N., Domschke, K., Eriksson, E., Fineberg, N. A., & Hättenschwiler, J. (2023). World Federation of Societies of Biological Psychiatry (WFSBP) guidelines for treatment of anxiety, obsessive-compulsive and posttraumatic stress disorders–Version 3. Part I: Anxiety disorders. *The World Journal of Biological Psychiatry*, *24*(2), 79-117.

Carter, C. S. (2022). Oxytocin and love: myths, metaphors and mysteries. *Comprehensive Psychoneuroendocrinology*, *9*, 100107.

Chaipunko, S., Sookkua, T., Nopparat, C., & Chutabhakdikul, N. (2025). Oxytocin Protects Against Corticosterone-Induced DA Dysfunction: An Involvement of the PKA/CREB Pathway. *Neurochemical Research*, *50*(1), 38.

DeMartini, J., Patel, G., & Fancher, T. L. (2019). Generalized anxiety disorder. *Annals of internal medicine*, *170*(7), ITC49-ITC64.

Gully, B. J., Brown, Z. E., Hornbacher, R., Brown, J. C., Back, S. E., McCance-Katz, E. F., Swift, R. M., & Haass-Koffler, C. L. (2024). Oxytocin reduces noradrenergic-induced opioid-like withdrawal symptoms in individuals on opioid agonist therapy. *Biological Psychiatry Global Open Science*, 100395.

Hirasawa, M., Kombian, S., & Pittman, Q. (2001). Oxytocin retrogradely inhibits evoked, but not miniature, EPSCs in the rat supraoptic nucleus: role of N-and P/Q-type calcium channels. *The Journal of physiology*, *532*(Pt 3), 595.

Hoge, E. A., Ivkovic, A., & Fricchione, G. L. (2012). Generalized anxiety disorder: diagnosis and treatment. *Bmj*, *345*.

Ivanov, S., Rashevskaya, T., & Makhonina, M. (2011). Flaxseed additive application in dairy products production. *Procedia Food Science*, *1*, 275-280.

Kaplan, H., Sadock, B., & Sadock, V. (2007). Synopsis of psychiatry behavioral sciences/clinical psychiatry, translated by Farzin Rezaei, 2ndVolume, Tehran. In: Arjmand publications.(persian).

Lecová, M., Babjáková, D., Sopková, D., Andrejčáková, Z., Hertelyová, Z., Petrilla, V., Polláková, M., & Vlčková, R. (2024). Different Lengths of Diet Supplementation with 10% Flaxseed Alter the Hormonal Profile and the Follicular Fluid Fatty Acid Content of Fattening Gilts. *Life*, *14*(2), 240.

Motovaseliyan, M., ghafari, F., & Kakhaki, M. T. (2016). Words equivalent to anxiety in Iranian traditional medicine texts. *Journal of Medical-Scientific Research History*, *8*(27), 47-72.

Mueed, A., Shibli, S., Korma, S. A., Madjirebaye, P., Esatbeyoglu, T., & Deng, Z. (2022). Flaxseed bioactive compounds: Chemical composition, functional properties, food applications and health benefits-related gut microbes. *Foods*, *11*(20), 3307.

Munk, M., Villalobo, E., Villalobo, A., & Berchtold, M. W. (2022). Differential expression of the three independent CaM genes coding for an identical protein: Potential relevance of distinct mRNA stability by different codon usage. *Cell Calcium*, *107*, 102656.

Oomah, B. D. (2001). Flaxseed as a functional food source. *Journal of the Science of Food and Agriculture*, *81*(9), 889-894.

Pal, S. K., & Shukla, Y. (2003). Herbal medicine: current status and the future. *Asian pacific journal of cancer prevention*, *4*(4), 281-288.

Rabow, S., Jonsson, H., Bro, E., & Olofsson, P. (2023). Cardiovascular effects of oxytocin and carbetocin at cesarean section. A prospective double-blind randomized study using noninvasive pulse wave analysis. *The Journal of Maternal-Fetal & Neonatal Medicine*, *36*(1), 2208252.

Rezaei-Tavirani, M., Razzaghi, Z., & Arjmand, B. (2022). Cholesterol metabolism pathway, the main target of coffee. *Research Journal of Pharmacognosy*, *9*(4), 39-47.

Service, U. o. W.-.-E. C. E., Crops, U. o. M. C. f. A., Products, & Service, M. E. (1990). *Alternative Field Crops Manual*. University of Wisconsin-Extension, Cooperative Extension. https://books.google.com/books?id=Sw6HOwAACAAJ

Shao, L., & Zhang, B. (2013). Traditional Chinese medicine network pharmacology: theory, methodology and application. *Chinese journal of natural medicines*, *11*(2), 110-120.

Singh, K., Mridula, D., Rehal, J., & Barnwal, P. (2011). Flaxseed: a potential source of food, feed and fiber. *Critical reviews in food science and nutrition*, *51*(3), 210-222.

Sirotkin, A. V. (2023). Influence of flaxseed (Linum usitatissimum) on female reproduction. *Planta Medica*, *89*(06), 608-615.

Tang, T., Zhang, X., Liu, Y., Peng, H., Zheng, B., Yin, Y., & Zeng, X. (2023). Machine learning on protein–protein interaction prediction: models, challenges and trends. *Briefings in Bioinformatics*, *24*(2), bbad076.

Tang, Y.-B., Tang, L., Chen, B., Fan, M.-J., Chen, G.-J., Ou, Y.-N., Yang, F., & Wu, X.-Z. (2024). Intranasal oxytocin alleviates postsurgical pain and comorbid anxiety in mice: Participation of BK (Ca) channels in the hippocampus. *Neuropharmacology*, 110243.

Tou, J. C., Chen, J., & Thompson, L. U. (1999). Dose, timing, and duration of flaxseed exposure affect reproductive indices and sex hormone levels in rats. *Journal of Toxicology and Environmental Health, Part A*, *56*(8), 555-570.

Touré, A., & Xueming, X. (2010). Flaxseed lignans: source, biosynthesis, metabolism, antioxidant activity, bio-active components, and health benefits. *Comprehensive reviews in food science and food safety*, *9*(3), 261-269.

Uvnaes Moberg, K., Julius, H., Handlin, L., & Petersson, M. (2022). Sensory stimulation and oxytocin: their roles in social interaction and health promotion. In (Vol. 13, pp. 929741): Frontiers Media SA.

Vlčková, R., Sopková, D., Andrejčáková, Z., Lecová, M., Fabian, D., Šefčíková, Z., Seidavi, A., & Sirotkin, A. V. (2022). Dietary supplementation of flaxseed (Linum usitatissimum L.) alters ovarian functions of xylene-exposed mice. *Life*, *12*(8), 1152.

Watson, R. R., & Preedy, V. R. (2012). *Bioactive Food as Dietary Interventions for Diabetes: Bioactive Foods in Chronic Disease States*. Elsevier Science. https://books.google.com/books?id=QOAyH0ZJ0JcC