Network Pharmacology of Ajmalicine

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Abstract- Network pharmacology has emerged as a promising interdisciplinary approach that integrates molecular biology, pharmacology, bioinformatics, and systems biology to understand complex drug-disease interactions. Unlike the traditional "one drug, one target" model, network pharmacology explores how a single compound can modulate multiple disease-related targets. This study investigates the bioactive alkaloid Ajmalicine, a known antihypertensive agent, through a network pharmacology framework. Molecular structures were retrieved from the PubChem database and analyzed using SwissTargetPrediction to identify potential gene and protein targets. Additional disease-related gene associations were derived from databases such as SEA, MalaCards, OMIM, and DisGeNET. Venn diagram analysis using Venny 2.1.0 identified overlapping gene targets, while pathway enrichment and protein-protein interaction networks were explored using Reactome, STRING, and Cytoscape. The findings highlight Ajmalicine's interaction with α1-adrenergic receptors and CYP2D6, its role in vasodilation, improved cerebral circulation, and neuroprotection. This integrative approach supports the potential of natural compounds and traditional herbal formulations in modern drug discovery and development, particularly for complex and chronic diseases.

Keywords: Ajmalicine, Network Pharmacology, SwissTargetPrediction, SEA, DisGeNET, OMIM, MalaCards, Venn Diagram, Reactome, STRING, Cytoscape, Antihypertensive, Gene Targets, Protein Interaction Network.

INTRODUCTION

The work of molecular biology and genetics research has produced vast data sets that aided in paining new in-sights into drug dis auvery processes. A single medication can target several nodes in the illness netweek, according to Hopkin, the founder of Nerwork Pharmacology [1]

The identification of the illness node is another key function of network pharmacology. In addition to these, it lowers the attrition rate in the illness network and enhances the number of clinical candidates with potency [2].

Molecular pharmacology, biochemical biology, and bioinformatics are among the various disciplines that are integrated in network pharmacology [1,3].

Network pharmacology has attracted more attention because of its high clinical investigation success rate, less or less expensive tid-rdable tide effects, improved drug efficacy, regulation of the signaling pathway with multiple pla channels, and interaction with multiple genes and proteins that could be easily targeted causing the disease [4].

Around 40% of the recent pharmacological discoveries are supplied by net-work pharmacology rather than a magic bullet concept [4, 5].

Today's network pharmacology offers a unique chance to systematically examine the relationship between complex illnesses and herbal formulas as well as the molecular complexity of herbal formulas [6, 7].

Herbs used in traditional remedies have shown a best molecular match, which might elicit a more consistent network reaction than a single drug [8,9,].

Network-based methodologies are becoming more popular research tools in areas of new drug development. By using natural products as the primary compound responsible for drug synergism and cumulative activity, they aid in the understanding of novel treatments. Numerous herbal formulations employed in traditional medicine have demonstrated the effectiveness of these procedures [9,10].

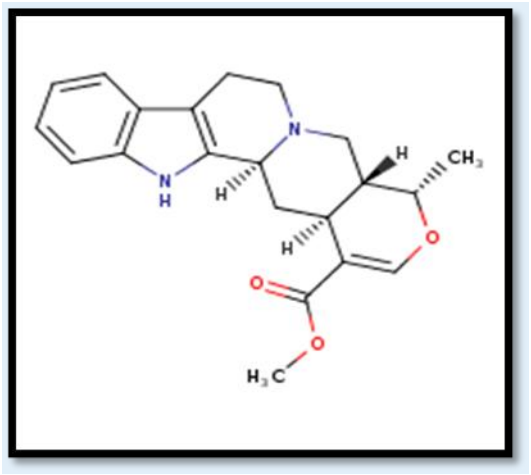
Herbs or herbal blends may be taken into consideration if their possibly active ingredients are recognized. Their application in herbal therapy served as the primary inspiration for this technique. Multi-drug targeted therapy and herbal formulations are comparable [11]. Selective polypharmacological techniques can easily employ active molecules to accomplish multi-target specific therapy [12,13].

This led to the development of network pharmacology, which integrates the linkages between medications and diseases as well as the interactions between drugs and

targets to uncover the mechanisms and pathways of pharmacological action. New treatment approaches have also been developed as a result of the shortcomings of current therapies for cancer, rheumatic, inflammatory, and chronic illnesses. Network pharmacology built on disease-related risks, driver genes, proteins, and drug targets can more successfully form disease signaling networks or modules than the conventional disease definition methods based on organs and symptoms. This helps to develop new and more effective treatment methods [14][15].

Advantage-Network pharmacology has several benefits, such as controlling the signaling pathway through various channels, improving therapeutic efficacy, lowering adverse effects, increasing clinical trial success rates, and lowering drug discovery expenses. Numerous genes and functional proteins interact in many complex illnesses.[16]

AJMALICINE-



Synonym-

δ-yohimbine, raubasine[17]

IUPACName-

methyl(19α)-19-methyl-16,17-didehydro-18-oxayohimban-16-carboxylate

Molecular Weight-
C21H24N2O3
Family-
Apocynaceae
Routes of administration-
oral
Average Mass- 352.434[17]

Ajmalicine is an antihypertensive medication used to treat excessive blood pressure. It is often referred to as raubasine or δ-yohimbine. [18] Card-Lamuran, Circolene, Cristanyl, Duxil, Duxor, Hydroxysarpon, Iskedyl, Isosarpan, Isquebral, Lamuran, Melanex, Raunatin, Saltucin Co, Salvalion, and Sarpan are only a few of the brand names under which it has been sold. [18] Numerous plants, including Rauvolfia spp., Catharanthus roseus, and Mitragyna speciosa, naturally contain this alkaloid. [18] [19] [20].

The drug, rauwolscine, and other yohimban derivatives share structural similarities with ajmalicine. [21] It has hypotensive rather than hypertension effects because, like corynanthine, it functions as an antagonist of α1-adrenergic receptors with preferred activities over α2-adrenergic receptors. [18,22] Furthermore, it is a potent inhibitor of the liver enzyme CYP2D6, which breaks down a lot of medications. 3.30 nM is its binding affinity at this receptor.

The significance of ajmalicine –

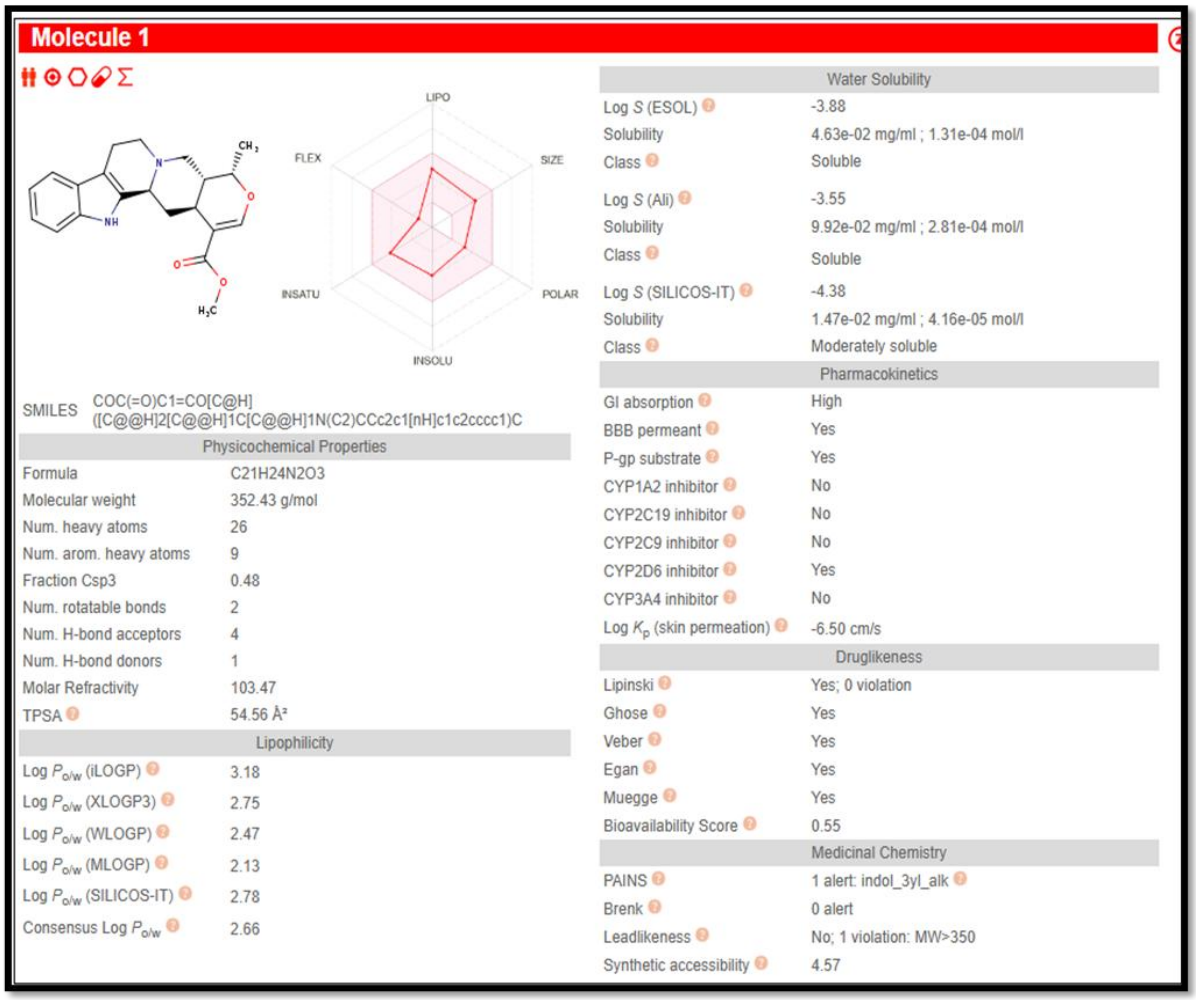
1. Antihypertensive Activity: Ajmalicine's vasodilatory qualities, which relax vascular smooth muscles, aid in decreasing blood pressure. Compared to reserpine, it is less effective but has fewer adverse effects.[23]
2. Improvement of Cerebral Circulation: Ajmalicine helps the brain receive more blood, which is why it is used to treat ailments like vertigo and memory problems, particularly in older individuals.
3. Neuroprotective Effects: Research has suggested anti-inflammatory and neuroprotective qualities that may be helpful in neurodegenerative diseases. [24]
4. Traditional Use: Rauvolfia serpentina, the source of ajmalicine, has been used for generations to treat mental illnesses, sleeplessness, and hypertension in Ayurvedic and traditional medical systems.[25]

MATERIAL AND METHODS

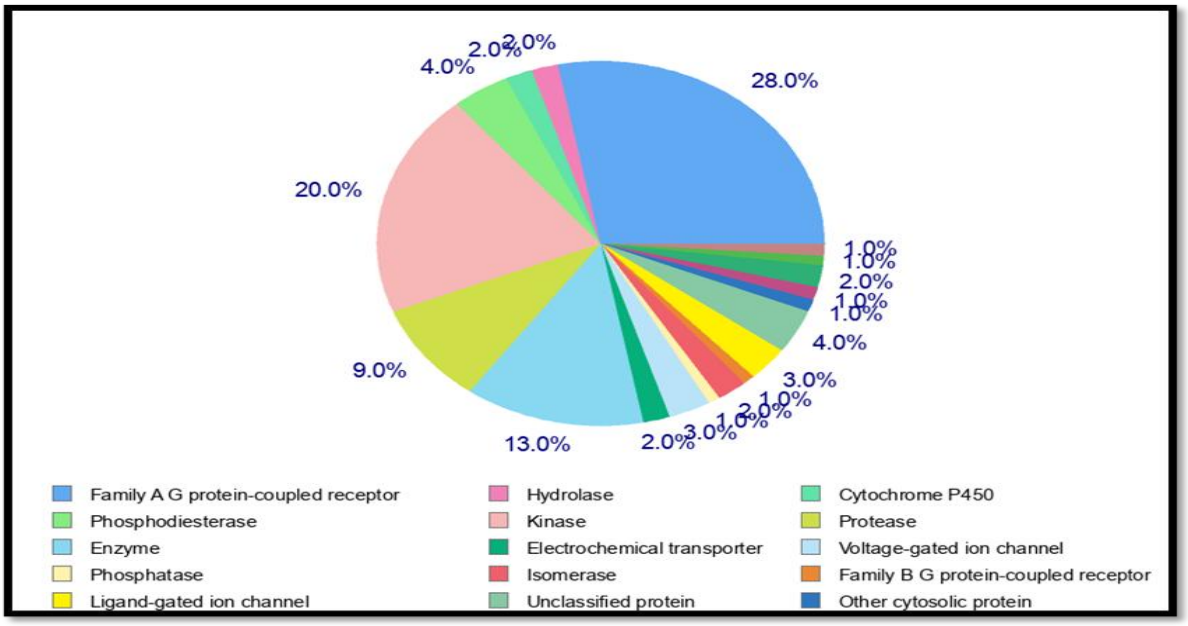
1)Selection of Phytochemical-

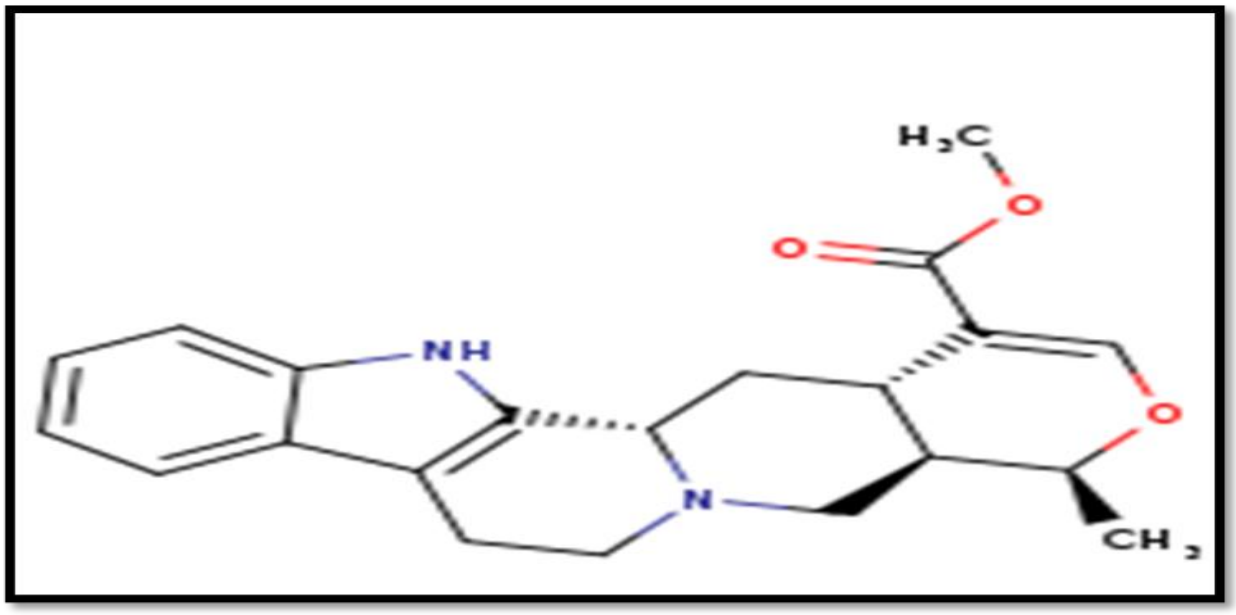
SMILIES OF AJMALICINE-
C[C@H]1[C@H]2CN3CCC4=C([C@@H]3C[C@@H]2C(=CO1)C(=O)OC)NC5=CC=CC=C45

SWISS ADME -



2) Selection of Target Identification –
SWISS TARGET





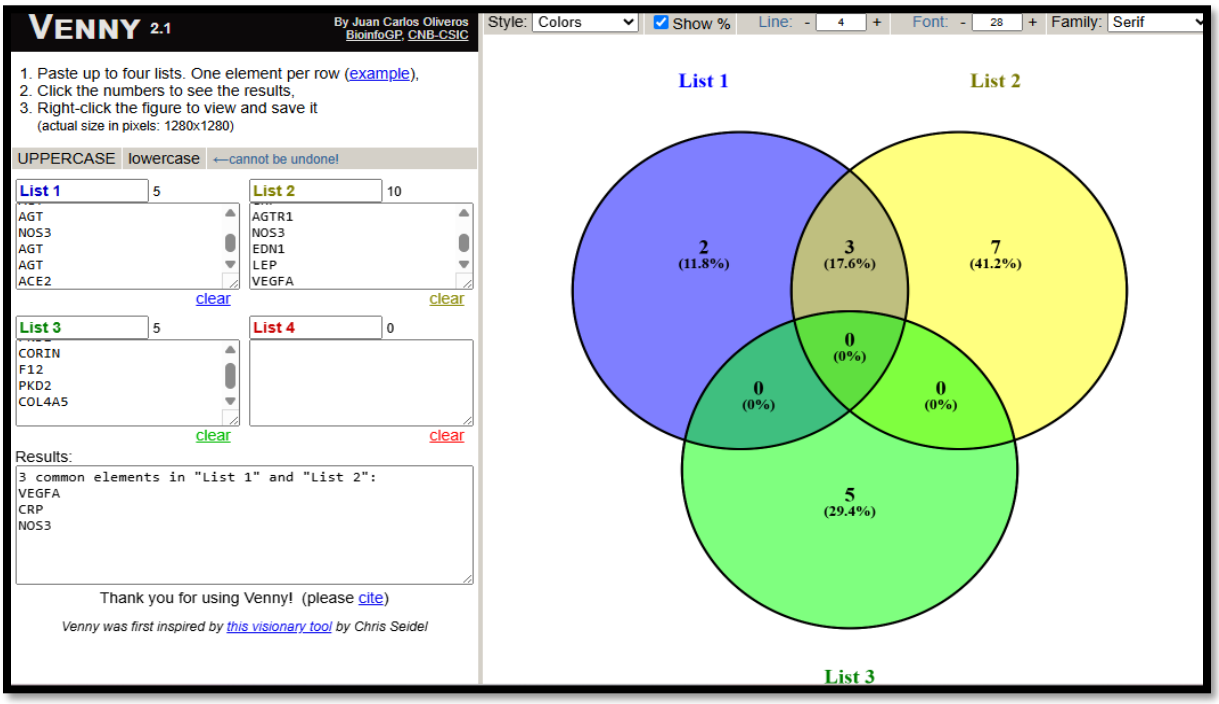
3)DISEASE GENES-
DISGENET and MALA CARDS-

DISGENET	MalaCards
VEGFA	PKD1
CRP	CORIN
AGT	F12
AGT	PKD2
AGT	COL4A5
AGT	MEN1
NOS3	DARS2
AGT	CELA2A
AGT	LOC129992813
ACE2	MTX2
ACE	

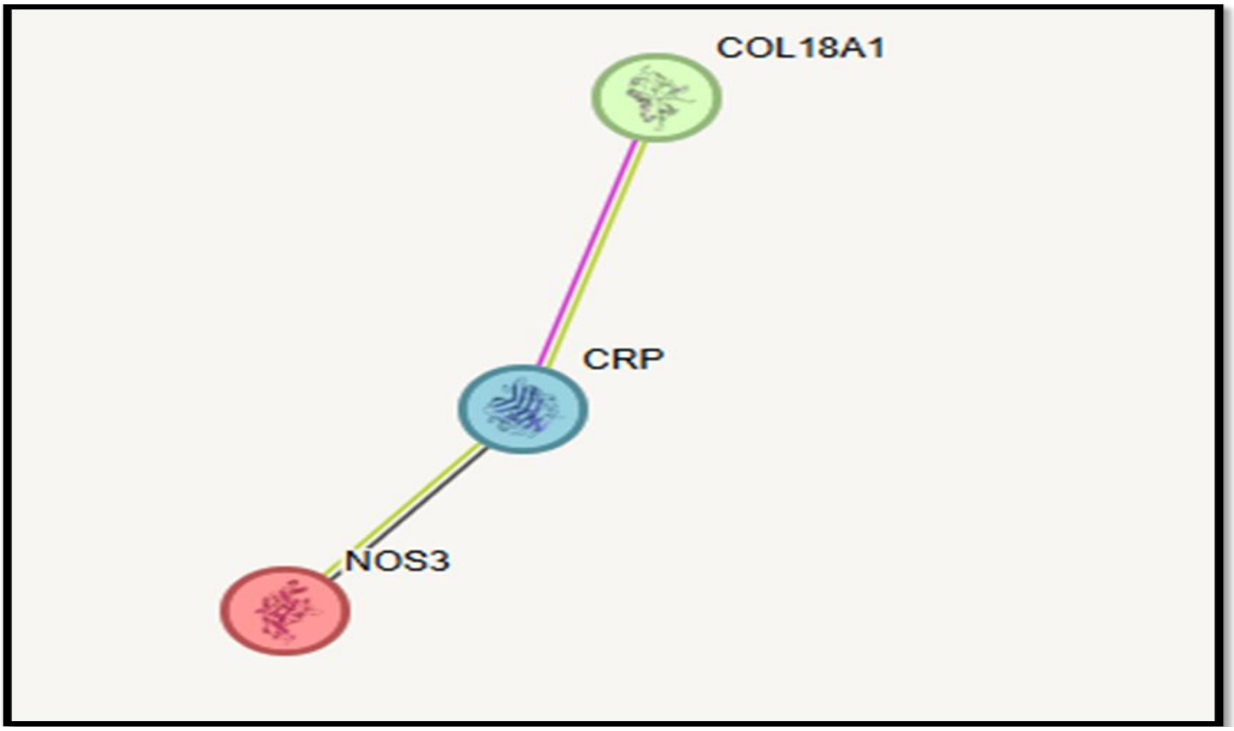
Gene	Gene Full Name	N diseases$_g$	N variants$_g$	Score$_{gda}$	N PMIDs
AGT	angiotensinogen	1230	99	1	1236
ACE	angiotensin I converting enzyme	1764	198	1	1201
INS	insulin	3105	79	1	701
REN	renin	821	51	1	606
CRP	C-reactive protein	3137	12	1	379
AGTR1	angiotensin II receptor type 1	618	79	1	254
NOS3	nitric oxide synthase 3	1003	45	1	226
EDN1	endothelin 1	1176	24	1	193
LEP	leptin	1667	90	1	168
VEGFA	vascular endothelial growth fact...	3131	35	1	163

4) Selection Of Common Gene –
VENNY

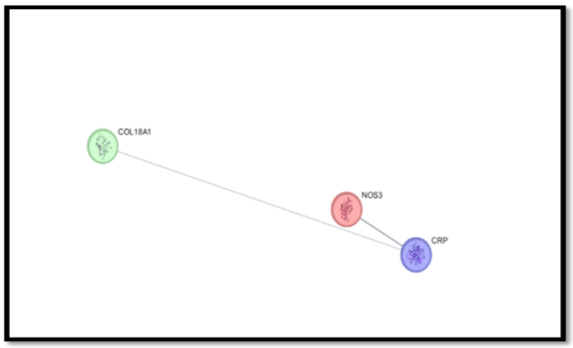
1. VEGFA
2. CRP
3. NOS3

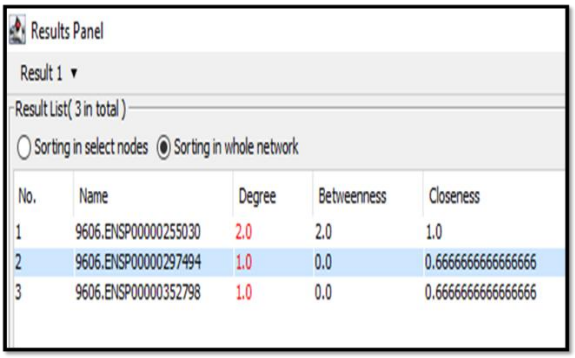


5) PPI Intraction and ranking-
STRING DATA-



6)CYTOSCAPE- CENTRALITY MEASURES

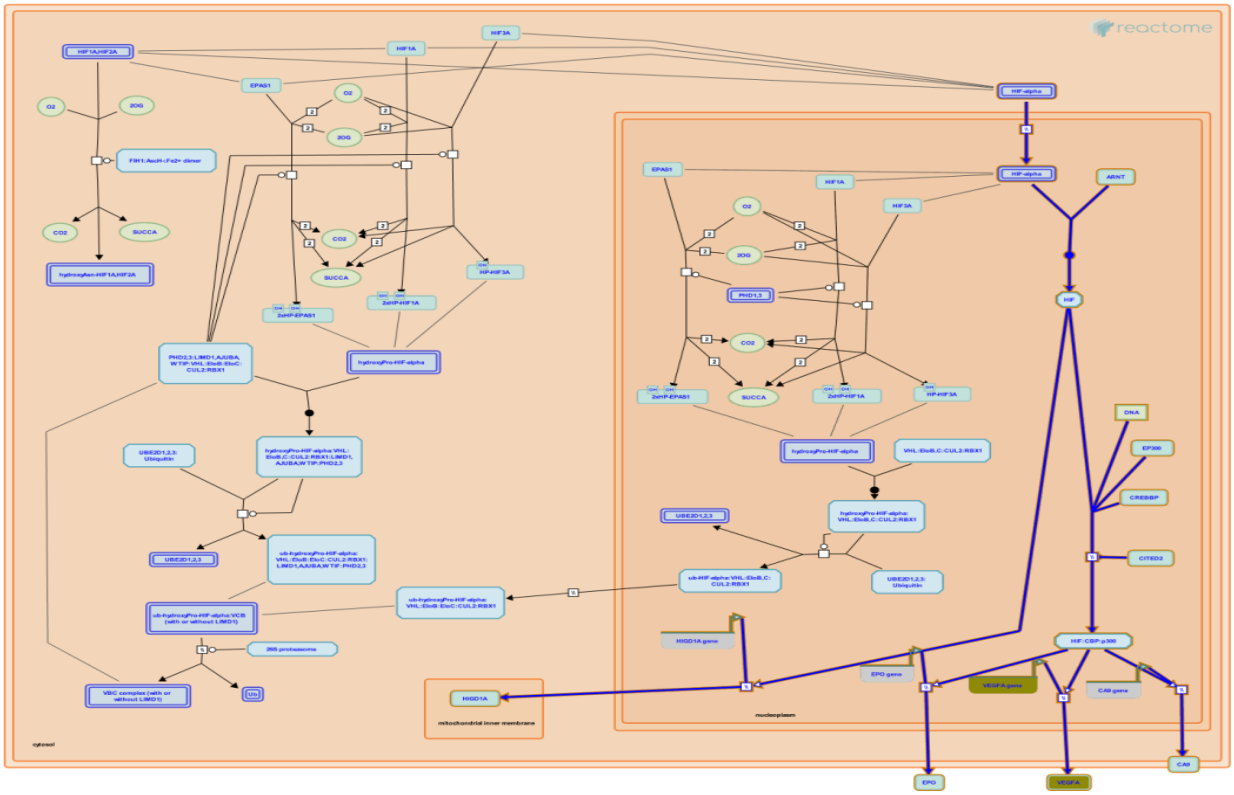




No.	Name	Degree	Betweenness	Closeness
1	9606.ENSP00000255030	2.0	2.0	1.0
2	9606.ENSP00000297494	1.0	0.0	0.6666666666666666
3	9606.ENSP00000352798	1.0	0.0	0.6666666666666666

RESULT-Centrality

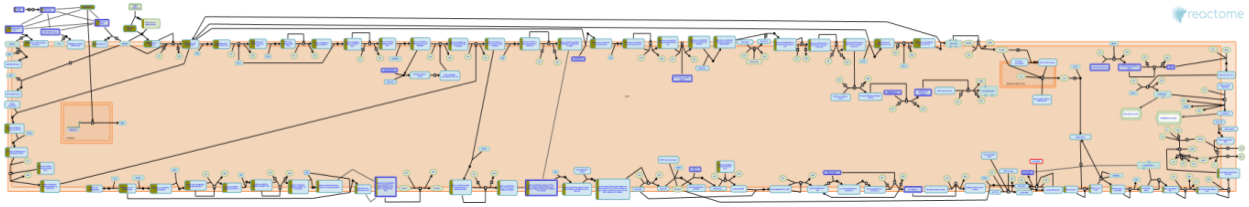
7) PATHWAY ANALYSIS REPORT-
REACTOME-
PATHWAT DETAILS-
1.Regulation of gene expression by Hypoxia-inducible Factor (R-HSA-1234158)



Cellular compartments: nucleoplasm.

HIF-alpha (HIF1A, HIF2A (EPAS1), HIF3A) is translocated to the nucleus, possibly by two pathways: importin 4/7 (Chachami et al. 2009) and importin alpha/beta (Depping et al. 2008). Once in the nucleus HIF-alpha heterodimerizes with HIF-beta (ARNT) (Wang et al. 1995, Jiang et al. 1996, Tian et al. 1997, Gu et al. 1998, Erbel et al. 2003) and recruits CBP and p300 to promoters of target genes (Ebert and Bunn 1998, Kallio et al. 1998, Ema et al. 1999, Gu et al. 2001, Dames et al. 2002, Freedman et al. 2002).[26]

2.Signaling by VEGF (R-HSA-194138)

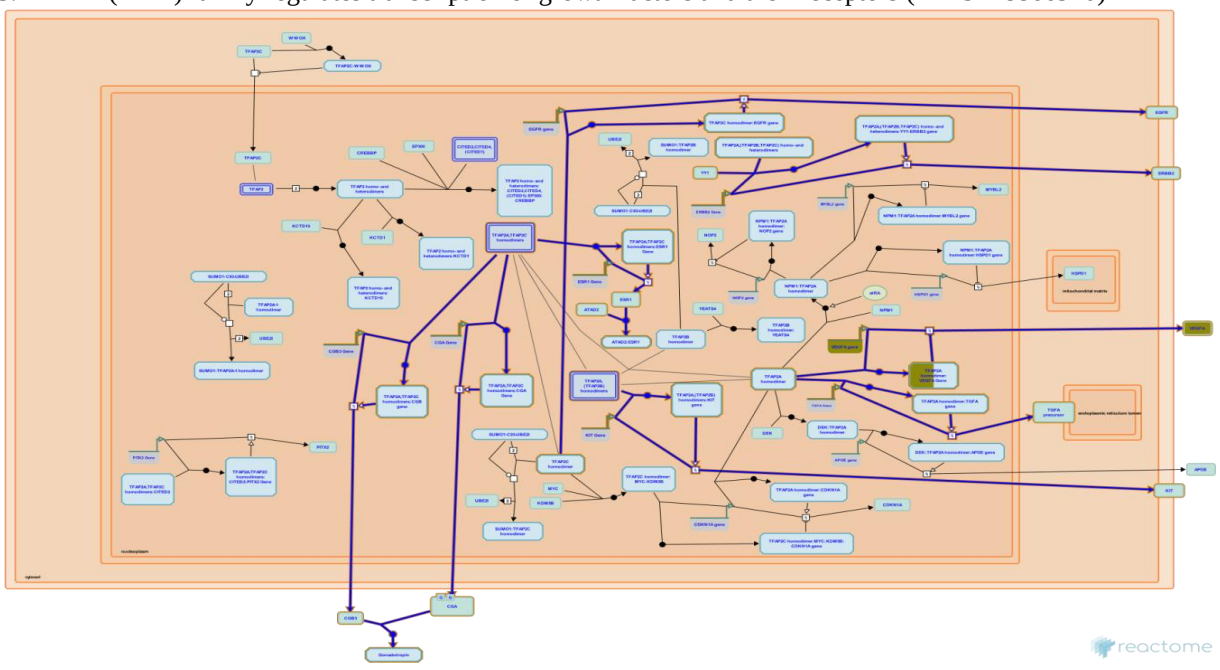


In normal development vascular endothelial growth factors (VEGFs) are crucial regulators of vascular development during embryogenesis (vasculogenesis) and blood-vessel formation in the adult (angiogenesis). In tumor progression, activation of VEGF pathways promotes tumor vascularization, facilitating tumor growth and metastasis. Abnormal VEGF function is also associated with inflammatory diseases including atherosclerosis, and hyperthyroidism. The members of the VEGF and VEGF-receptor protein families have distinct but overlapping ligand-receptor specificities, celltype expression, and function. VEGF-receptor activation in turn regulates a network of signaling processes in the body that promote endothelial cell growth, migration and survival (Hicklin and Ellis, 2005; Shibuya and Claesson-Welsh, 2006).

Molecular features of the VGF signaling cascades are outlined in the figure below (from Olsson et al. 2006; Nature Publishing Group). Tyrosine residues in the intracellular domains of VEGF receptors 1, 2,and 3 are indicated by dark blue boxes; residues susceptible to phosphorylation are numbered. A circled R indicates that phosphorylation is regulated by cell state (VEGFR2), by ligand binding (VEGFR1), or by heterodimerization (VEGFR3). Specific phosphorylation sites (boxed numbers) bind signaling molecules (dark blue ovals), whose interaction with other cytosolic signaling molecules (light blue ovals) leads to specific cellular (pale blue boxes) and tissue-level (pink boxes) responses in vivo. Signaling cascades whose molecular details are unclear are indicated by dashed arrows. DAG, diacylglycerol; EC, endothelial cell; eNOS, endothelial nitric oxide synthase; FAK, focal adhesion kinase; HPC, hematopoietic progenitor cell; HSP27, heat-shock protein-27; MAPK, mitogen-activated protein kinase; MEK, MAPK and ERK kinase; PI3K, phosphatidylinositol 3' kinase; PKC, protein kinase C; PLCgamma, phospholipase C-gamma; Shb, SH2 and beta-cells; TSAd, T-cellspecific adaptor.

In the current release, the first events in these cascades - the interactions between VEGF proteins and their receptors - are annotated.[27]

3. TFAP2 (AP-2) family regulates transcription of growth factors and their receptors (R-HSA-8866910)



TFAP2A and TFAP2C directly stimulate transcription of the estrogen receptor ESR1 gene (McPherson and Weigel 1999). TFAP2A expression correlates with ESR1 expression in breast cancer, and TFAP2C is frequently overexpressed in estrogen-positive breast cancer and endometrial cancer (deConinck et al. 1995, Turner et al. 1998). TFAP2A, TFAP2C, as well as TFAP2B can directly stimulate the expression of ERBB2, another important breast cancer gene (Bosher et al. 1996). Association of TFAP2A with the YY1 transcription factor significantly increases the ERBB2 transcription rate (Begon et al. 2005). In addition to ERBB2, the expression of another receptor tyrosine kinase, KIT, is also stimulated by TFAP2A and TFAP2B (Huang et al. 1998), while the expression of the VEGF receptor tyrosine kinase ligand VEGFA is repressed by TFAP2A (Ruiz et al. 2004, Li et al. 2012). TFAP2A stimulates transcription of the transforming growth factor alpha (TGFA) gene (Wang et al. 1997). TFAP2C regulates EGFR expression in luminal breast cancer (De Andrade et al. 2016). In placenta, TFAP2A and TFAP2C directly stimulate transcription of both subunits of the human chorionic gonadotropin, CGA and CGB (Johnson et al. 1997, LiCalsi et al. 2000).[28]

RESULT AND DISSCUSSION

- In the presence investigation, genes associated with the lead and receptor molecules were analyzed.
- The new study also uses the Pubchem to determine the grins of chemicals. By incorporating these smiles into the complex procedure called as "swiss target prediction," uniport ids for the genes and receptors were obtained.
- The present study then found that additional genes were extracted from the A Ajmalicine molecule and from the SEA tool using a number of tools, such as Swiss target prediction, Malacards, Omim, and Disgenet. Additionally, an Al tool was used to identify common genes.
- The common gene was obtained using Venny 2.1.0.
- In the current Network Pharmacology investigation, a string tool from Cytoscape was used for centrality measurements, and Reactome software was used to obtain disease pathways.
- Therefore, network pharmacology provides fresh understanding into drug action study.

REFERENCE

[1] Hopkins, A. L. “Network Pharmacology: The next Paradigm in Drug Discovery.” Nature Chemical Biology, no. 4, 2008, pp. 682–690.

[2] Hopkins, A. L. “Network Pharmacology.” Nature Biotechnology, vol. 25, no. 10, 2007, pp. 1110–1110.

[3] Roth, Bryan L., et al. “Magic Shotguns versus Magic Bullets: Selectively Non-Selective Drugs for Mood Disorders and Schizophrenia.” Nature Reviews. Drug Discovery, vol. 3, no. 4, 2004, pp. 353–359, doi:10.1038/nrd1346.

[4] Wu, X.; Jiang, R.; Zhang, M.Q.; Li, S., Network-based global inference of human disease genes. Molecular systems biology, 2008, 4, (1), 189.

[5] Zhang, G., et al. Network Pharmacol-Ogy: A New Approach for Chinese Herbal Medicine Research. Evi-Dence-Based Complementary and Alternative Medicine. 2013.

[6] Zuo, Jian, et al. “Integrating Network Pharmacology and Metabolomics Study on Anti-Rheumatic Mechanisms and Antagonistic Effects against Methotrexate-Induced Toxicity of Qing-Luo-Yin.” Frontiers in Pharmacology, vol. 9, 2018, p. 1472, doi:10.3389/fphar.2018.01472.

[7] Yang, Haixing, et al. “A Novel Systems Pharmacology Model for Herbal Medicine Injection: A Case Using Reduning Injection.” BMC Complementary and Alternative Medicine, vol. 14, no. 1, 2014, p. 430, doi:10.1186/1472-6882-14-430.

[8] Hao, Haiping, et al. “Insights into Drug Discovery from Natural Medicines Using Reverse Pharmacokinetics.” Trends in Pharmacological Sciences, vol. 35, no. 4, 2014, pp. 168–177, doi:10.1016/j.tips.2014.02.001.

[9] Emig, Dorothea, et al. “Drug Target Prediction and Repositioning Using an Integrated Network-Based Approach.” PloS One, vol. 8, no. 4, 2013, p. e60618, doi:10.1371/journal.pone.0060618.

[10] Shahreza, Lotfi, et al. “A Review of Network-Based Approaches to Drug Repositioning.” Brief. Bioinform, vol. 19, 2018, pp. 878–892.

[11] Hao, D. C., and P. G. Xiao. "Network Pharmacology: A Rosetta Stone for Traditional C Hinese Medicine." Drug Dev. Res, vol. 75, 2014

[12] Morphy, Richard, et al. "From Magic Bullets to Designed Multiple Ligands." Drug Discovery Today, vol. 9, no. 15, 2004, pp. 641–651, doi:10.1016/S1359-6446(04)03163-0

[13] Morphy, Richard, et al. "From Magic Bullets to Designed Multiple Ligands." Drug Discovery Today, vol. 9, no. 15, 2004, pp. 641–651, doi:10.1016/S1359-6446(04)03163-0.

[14] C., et al. "Schmidt Network Pharmacology: Curing Causal Mechanisms Instead of Treating Symptoms Trends Pharmacol." Trends Pharmacol. Sci, vol. 43, 2022, pp. 136–150, doi:10.1016/j.tips.2021.11.004.

[15] Li, X., et al. "Fan Network Pharmacology Approaches for Research of Traditional Chinese Medicines Chin." J. Nat. Med, vol. 21, 2023, pp. 323–332, doi:10.1016/s1875-5364(23)60429-7.

[16] Zhang, G., Li, Q., Chen, Q., & Su, S. (2013). Network Pharmacology: a new approach for Chinese herbal medicine research. Evidence-based Complementary and Alternative Medicine, 2013, 1–9. https://doi.org/10.1155/2013/621423.

[17] Brown, Richard T., et al. "Conversion of Secologanin into Elenolic Acid and 18-Oxayohimban Alkaloids." Journal of the Chemical Society, Perkin Transactions 1, no. 2, 1976, p. 160, doi:10.1039/p19760000160.

[18] Roberts, Margaret F. Alkaloids: Biochemistry, Ecology, and Medicinal Applications. Springer Science & Business Media, 1998

[19] Kurz, W. G., et al. "Alkaloid Production in Catharanthus Roseus Cell Cultures VIII." Planta Medica, vol. 42, no. 1, 1981, pp. 22–31, doi:10.1055/s-2007-971541

[20] León, F., et al. "Phytochemical Characterization of the Leaves of Mitragyna Speciosa Grown in U." Natural Product Communications, vol. 4, no. 7, 2009, pp. 907–910, doi:10.1177/1934578X0900400705.PMC9255435

[21] Roberts, Margaret F. Alkaloids: Biochemistry, Ecology, and Medicinal Applications. Springer Science & Business Media, 1998

[22] Strobl, G. R., et al. "Development of a Pharmacophore for Inhibition of Human Liver Cytochrome P-450 2D6: Molecular Modeling and Inhibition Studies." Journal of Medicinal Chemistry, vol. 36, no. 9, 1993, pp. 1136–1145, doi:10.1021/jm00061a004.

[23] Dey, Lucy, et al. "Alternative Therapies for Type 2 Diabetes." Alternative Medicine Review: A Journal of Clinical Therapeutic, vol. 7, no. 1, 2002, pp. 45–58

[24] Dutta, N. K., et al. "The Anti-Inflammatory Potential of Ajmalicine." Indian Journal of Experimental Biology, vol. 46, no. 7, 2008, pp. 555–560.

[25] Yang, Jing-Yi, et al. "Characterization and Comparison Analysis of WHO Monographs on Selected Medicinal Plants and ISO Standards of Chinese Materia Medica." Chinese Journal of Integrative Medicine, vol. 29, no. 6, 2023, pp. 540–548, doi:10.1007/s11655-022-3526-7.

[26] Semenza GL, Rue EA, Wang GL & Jiang BH (1995). Hypoxia-inducible factor 1 is a basic-helix-loop-helix-PAS heterodimer regulated by cellular 02 tension, Proc Natl Acad Sci USA, 92, 5510-4.

[27] Matsumoto T, Cross MJ, Dixelius J & Claesson-Welsh 1. (2003). VEGF-receptor signal transduction. Trends Biochem Sci, 28, 488-94.

[28] Wang D, Shin TH & Kudlow JE (1997). Transcription factor AP-2 controls transcription of the hu-man transforming growth factor-alpha gene. J. Biol. Chem., 272, 14244-50,