



Evaluation of Verteporfin as a Photosensitizer With Anticancer Activity Via System Biology Tools

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Received: February 24, 2025

Accepted: June 3, 2025

ePublished: August 18, 2025

Abstract

Introduction: Verteporfin as a photosensitizer in photodynamic therapy (PDT) is used to inhibit deviant choroidal vascularization. This agent also has an anticancer property. The effective treatment of various tumors such as melanoma by using verteporfin is confirmed. In the present study, the ability of verteporfin in growth inhibition of MKN45 is investigated via protein-protein interaction (PPI) analysis to explore its beneficial role in PDT.

Methods: The gene expression profiles of the treated MKN45 cell with verteporfin are mined from the Gene Expression Omnibus (GEO) database and evaluated via PPI network analysis to find the central genes. The central genes are searched in the GeneCards database to find the more related genes to gastric cancer. The expression amounts of the top central genes related to stomach adenocarcinoma are extracted from the University of Alabama at Birmingham Cancer data analysis Portal (UALCAN).

Results: Among 2356 significant differentially expressed genes (DEGs), 58 hub-bottlenecks were determined via PPI network analysis. ERBB2, EGFR, CREBBP, CTSD, PSEN1, HRAS, PIK3R1, NRAS, NFKBIA, FOS, CTSD, CALR, NFE2L2, RHOA, CDKN1A, ITGB1, APP, CD44, HSPA5, and HMGR were pointed out as the crucial genes. The expression amounts of ERBB2, EGFR, CREBBP, CTSD, and PSEN1 genes which were highlighted as critical genes via the directed PPI network were explored for stomach adenocarcinoma.

Conclusion: In conclusion, the anticancer property of verteporfin was highlighted. This finding can improve the efficacy of related PDT.

Keywords: Verteporfin, Photosensitizer, Anticancer, Gene expression, Network analysis



Introduction

Photodynamic therapy (PDT) is an attractive feature of light application in medicine. Experts highlight the role of light and various types of photosensitizers in PDT as a promising approach to treating a wide range of diseases. The interaction between light and a photosensitizer in the presence of molecular oxygen has made PDT a repeatable, non-toxic, and noninvasive method for treating tumors and promoting wound healing¹ PDT displays the unresolved presentation in cancer therapy, but some unavoidable disadvantages should be solved. Limitations of PDT to treat solid tumors appear as poor tumor selectivity, limited light penetration depth, and dependency on oxygen presence. Discovering new, efficient photosensitizers, along with other efforts and

protocols, has gone into overcoming these complications.² Photosensitizers are the molecules that, after receiving an explicit light wavelength, create reactive oxygen species via reaction with the molecular oxygen. The result of this reaction leads to cell death in targeted tissue.³ Verteporfin is a photosensitizer that has been administrated in PDT to inhibit deviant choroidal vascularization. This method is used as a safe and effective treatment for patients with choroidal neovascularization. A combination of this therapeutic tool with VEGF inhibition results in effective treatment in patients.⁴ It is reported that verteporfin has an anticancer property besides photosensitizer charisma. Jeong SB et al. published data about the anticancer activity of verteporfin versus non-small cell lung cancer via the downregulation of ANO1.⁵ Kang et al have investigated

the effect of verteporfin on gastric cancer cell growth. Based on this document, verteporfin inhibited the studied cell growth via overwhelming adhesion molecule FAT1.⁶ Investigations indicate that hyaluronic acid nanoparticle/ verteporfin is used as an operative nanomedicine for the treatment of uveal melanoma in a mouse model.⁷

Gastric cancer is known as an important malignancy with high load lethality and disease in the world. Targeted therapy, chemotherapy, and immunotherapy have been developed to treat gastric cancer.^{8,9} Gene expression analysis as an attractive method is used to evaluate the treatment efficacy of many drugs and methods. However, the results of such experiments have been interpreted via bioinformatics. Protein-protein interaction (PPI) network analysis is a well-known method for analyzing genomics findings. Genes are included in an interactome to form a PPI network via edges. The central nodes of the PPI network, such as the hub genes, play a critical role in the related functions of the network.¹⁰⁻¹² Nejad MR et al. have studied laser intensity effects on PDT efficacy via PPI network analysis. The effect of photosensitizer concentration on the efficacy of PDT has been studied by Vafaei et al.^{13,14} In the present study, the gene expression profiles of gastric cancer MKN45 cell in the presence of verteporfin is extracted from the GEO database and analyzed via the PPI network to highlight the photosensitizer property of this agent versus its anticancer character to inhibit the growth of gastric cancer cells. The combination of both anticancer and photosensitizer properties of verteporfin can be considered as a potential therapeutic method for decreasing the site effect and increasing the efficacy of treatment.

Methods

Data Collection and Pre-evaluation Analysis

Data were mined from the GEO database. Verteporfin (trade name Visudyne) is a photosensitizer medication that is used in PDT. The light wavelength of 689 nm stimulates this agent. This substance is applied to treat abnormal blood vessels as a photosensitizer via PDT¹⁵. Data of this gene set are related to the anticancer property of verteporfin on MKN45 cells. Data are associated with the publication of Kang et al.⁶ To examine the efficacy of verteporfin versus gastric cancer, the MKN45 gastric cancer cells were treated and their gene expression changes were compared with the cells in the presence of DMSO as control individuals. The gene expression profiles were studied via microarray. Data were analyzed via the GEO2R program to determine the significant differentially expressed genes (DEGs). A Uniform Manifold Approximation and Projection (UMAP) plot was applied to visualize the separation of the treated cells from controls. The uncharacterized DEGs were ignored from more assessments. The significant DEGs were identified based on an adjusted P value < 0.05 .

PPI Network Analysis

The significant DEGs were included in the “Protein query” of the STRING database via Cytoscape software to form an undirected PPI network. The PPI network was analyzed via the “Network Analyzer” application of Cytoscape software to find the hub genes. The hubs were determined based on a degree value cutoff of (mean + 2 (standard deviation)). The top 5% of genes were introduced as bottlenecks. The common hubs and bottlenecks were determined as hub-bottlenecks. The hub-bottleneck genes were assessed via a directed PPI network to screen the genes. The genes were included in the CluePedia plugin of Cytoscape software. The nodes were connected via activation, inhibition, expression, reaction, catalysis, and post-translation modification actions.

Gene Cards Assessment

The phrase “gastric cancer” was searched in the GeneCards database to find the related genes. The extracted genes were classified into seven calls of genes based on GIFtS value with an interval of 10. The top-ranked group of genes was selected based on high scores as gastric cancer-related genes. The hub-bottleneck DEGs from PPI network analysis were searched in this top group of the genes of GeneCards to find the common individuals.

The Cancer Genome Atlas (TCGA) Analysis

Expression of the 5 top gastric cancer-related hub-bottlenecks was mined from The University of Alabama at Birmingham CANcer data analysis Portal (UALCAN) (<https://ualcan.path.uab.edu/analysis.html>) linked with The Cancer Genome Atlas (TCGA) database.

Results

The UMAP plot analysis demonstrated the complete separation of the gene expression profiles of the treated AGS cells from the controls. As depicted in Figure 1, each

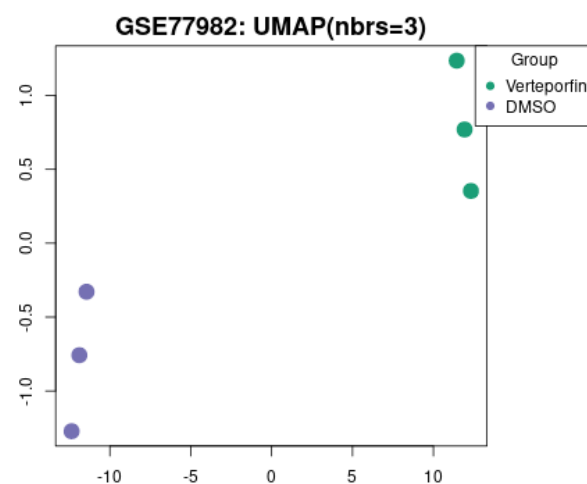


Figure 1. UMAP Plot of the Treated AGS Cells Via Photodynamic Therapy Versus the Individuals in the Presence of DMSO as Controls. Gene expression profiles of each sample is compared with three neighbors

profile is compared with 3 neighbors. The results indicated that the treated cells were separated from controlled individuals. The assessment showed that 2356 genes were dysregulated significantly. After cleaning the data (deleting the uncharacterized DEGs), 2120 individuals were selected for more investigation. Among the 2120 significant DEGs, 1778 individuals were recognized by the STRING database. The PPI network including 1778 nodes and 12 802 edges was constructed. Eighty-five hubs, 85 bottlenecks, and 58 hub-bottlenecks were identified as the main connected component of the network. The 58 hub-bottleneck genes were included in a directed PPI network. Among the 58 queried hub-bottlenecks, 43 individuals participated in the main connected component of the constructed directed PPI network (see Figure 2).

A total number of 7014 gastric cancer-related genes were mined from the GeneCards database. The genes were classified into seven groups (see Figure 3). As depicted in Figure 3, the top group includes 644 more related genes. There are 20 common genes between the hub-bottlenecks and the genes of top group from GeneCards (see Table 1). These 20 genes were highlighted as the more related ones to gastric cancer hub-bottlenecks. These 20 genes were searched among the directed PPI network (see Figure 2). The results including two isolated genes and a connected

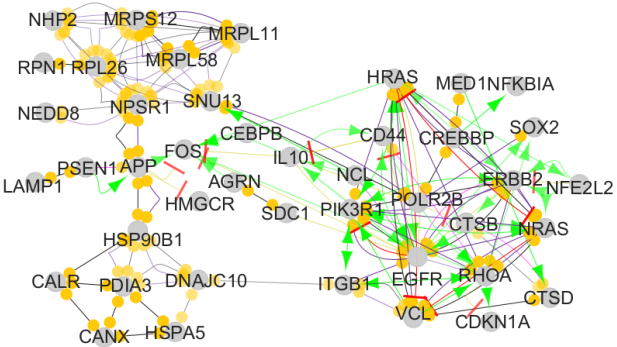


Figure 2. The Main Connected Component of the Directed PPI Network of the Queried Hub-Bottlenecks. Green, red, yellow, purple, pink, and black refer to activation, inhibition, expression, catalysis, post translation modification, and reaction, respectively

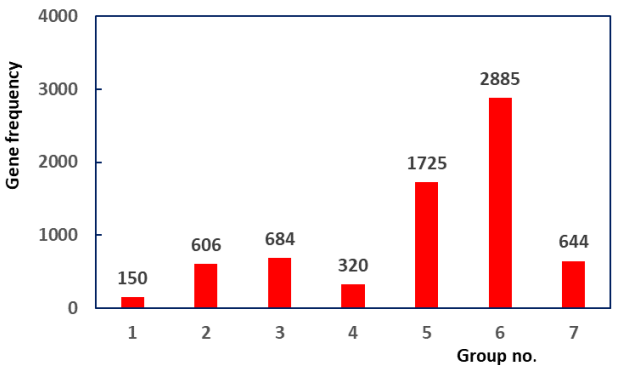


Figure 3. The Gastric Cancer Related Genes From GeneCards Database. The presented grouped are classified based on the GIFtS interval =10, From 0 to 70. Data are arranged from left to right ascendingly

component are presented in Figure 4. Gene expression amounts for ERBB2, EGFR, CREBBP, CTSD, and PSEN1 from TCGA in stomach adenocarcinoma relative to controls are demonstrated in Figure 5.

Discussion

The significant role of verteporfin as a photosensitizer is recorded in the literature.¹⁶ Twenty critical genes are highlighted as the targets of verteporfin in MKN45 cells. The involvement of the top five genes in cancer and the effect of verteporfin on their expression are discussed in the following part. Erb-b2 receptor tyrosine kinase 2 (ERBB2) (previously referred to as HER2) is a member of a family of epidermal growth factor receptors which also includes EGFRs of ERBB1, ERBB3, and ERBB4.¹⁷ Investigations indicate that the activation of the ERBB2 receptor signaling pathways is accompanied by an enhancement of various metastasis-associated properties. This effect is associated with an increase in cancer metastasis. Furthermore, the upregulation of ERBB2 leads to therapeutic resistance in patients.¹⁸ ERBB2 appears as a downregulated critical gene in our analysis. As depicted in Figure 5,

Table 1. The Common Hub-Bottleneck Genes With the Genes of the Top Group From GeneCards Database

No.	Gene	Rank in GeneCards Based on GIFtS	No.	Gene	Rank in GeneCards Based on GIFtS
1	ERBB2	3	11	CTSB	160
2	EGFR	4	12	CALR	162
3	CREBBP	24	13	NFE2L2	170
4	CTSD	59	14	RHOA	205
5	PSEN1	80	15	CDKN1A	211
6	HRAS	83	16	ITGB1	225
7	PIK3R1	102	17	APP	305
8	NRAS	119	18	CD44	454
9	NFKBIA	143	19	HSPA5	471
10	FOS	152	20	HMGCR	574

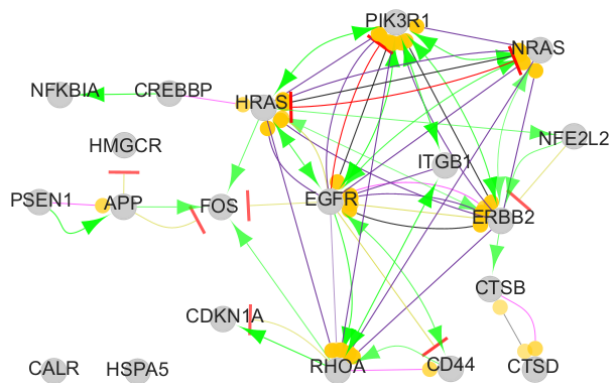


Figure 4. The 20 Common Hub-Bottleneck Genes With the Genes of the Top Group From GeneCards Database Searched in the Directed PPI in Figure 2. Green, red, yellow, purple, pink, and black refer to activation, inhibition, expression, catalysis, post translation modification, and reaction, respectively

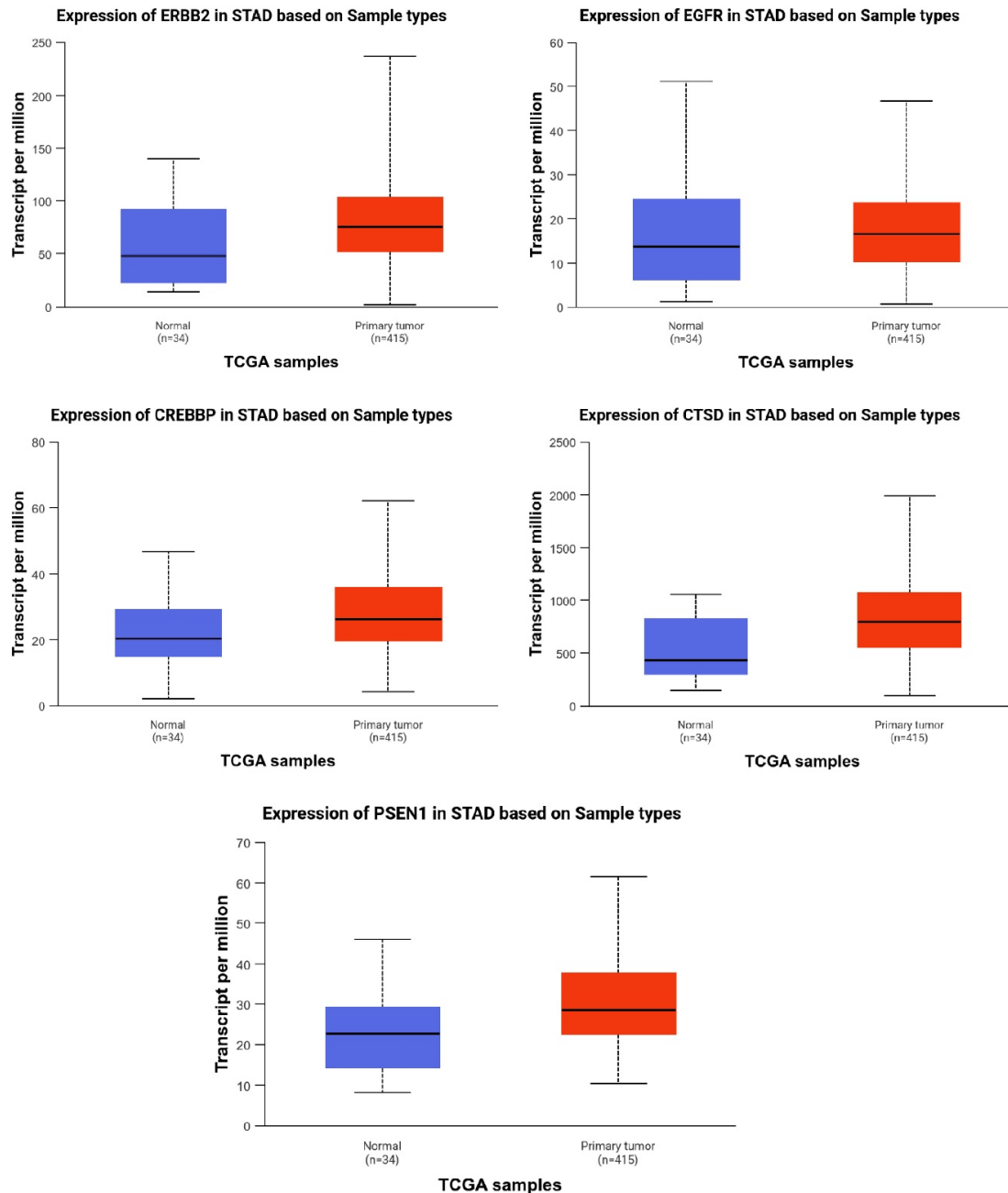


Figure 5. Gene Expression Amounts for ERBB2, EGFR, CREBBP, CTSD, and PSEN1 From TCGA in Stomach Adenocarcinoma (STAD) Relative to Controls. The median of data are comparable

ERBB2 is upregulated in stomach adenocarcinoma. As administrated in Table 1, ERBB2 is the top relevant gene to gastric cancer. The downregulation of ERBB2 in the treated MKN45 cells in the presence of verteporfin refers to the anticancer property of this photosensitizer agent.

The second downregulated critical gene is the EGFR. As demonstrated in Figure 5, EGFR is upregulated in stomach adenocarcinoma. Following ERBB2, EGFR is the most relevant gene to gastric cancer. Its GIFts and rank in GeneCards are 68 and 4, respectively. The overexpression of EGFR in certain cancer types has been reported by researchers. The involvement of EGFR in the regulation of the biological features of cancer progression, such as

metastasis, proliferation, and drug resistance, has been accentuated in the literature. Various medicines targeting EGFR inhibition appear as a suitable method for treating several types of cancer¹⁹. Positive co-expression between ERBB2 and EGFR is illustrated in Figure 4. Similar to the downregulation of ERBB2, the decrement of EGFR expression in the presence of verteporfin is clear evidence about its role in controlling the growth of the treated cancer cells.

The third downregulated critical hub-bottleneck gene is the CREB binding protein (CREBBP). On the basis of the literature, the upregulation and activation of cAMP response element-binding protein (CREB) is

accompanied by tumor growth.²⁰ The association between tumor volume and CREBBP expression in patients with Grade-3 glioma has been reported by experts.²¹ LeBlanc et al's investigation demonstrated that targeting CREBBP overwhelms chemotherapy resistance in patients with acute myeloid leukemia when they were treated with azacitidine and Venetoclax.²² TCGA assessment showed the upregulation of CREBBP in stomach adenocarcinoma (see Figure 5). The suppression of CREBBP in MKN45 cells highlights the antitumor property of verteporfin.

Cathepsin D (CTSD) is the fourth downregulated gene in the presence of verteporfin. There is a positive correlation between the excessive levels of CTSD outside lysosomes and the cell membrane and cancer development, invasion, migration, and angiogenesis. The association of human CTSD downregulation and neurodegenerative disorders has been reported in the literature.²³ As depicted in Figure 5, CTSD is upregulated in stomach adenocarcinoma. The downregulation of CTSD in the presence of verteporfin refers to the anticancer property of the studied photosensitizer. Presenilin1 (PSEN1) is the fifth crucial hub-bottleneck that is regulated under the effect of verteporfin in MKN45 cancer cells. This gene is studied frequently in neurodegenerative diseases such as the early onset of Alzheimer's disease, frontotemporal dementia, dementia with Lewy bodies, and Parkinson's disease.²⁴ As depicted in Table 1, the rank of PSEN1 in GeneCards is too far from the top three genes.

Conclusion

In conclusion, verteporfin inhibits MKN45 cancer cells by suppressing crucial genes such as ERBB2, EGFR, and CREBBP, the well-related genes to gastric cancer. Since verteporfin is a photosensitizer agent, this antitumor property of verteporfin makes it a powerful photosensitizer in PDT. This dual character of verteporfin has possible advantages such as the augmentation of efficacy in cancer therapy and the reduction of side effects. These findings open new windows about the application of verteporfin as a remarkable photosensitizer in the PDT of cancers.

Authors' Contribution

Conceptualization: Zahra Razzaghi, Fatemeh Bandarian.

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Supervision: Babak Arjmand.

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Writing—original draft: Zahra Razzaghi, Fatemeh Bandarian.

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Competing Interests

There is no conflict of interest between authors.

Ethical Approval

This project is approved by IR.SBMU.LASER.REC.1403.038 ethical code.

Funding

This project is supported by Shahid Beheshti University of Medical Sciences.

References

- Niculescu AG, Grumezescu AM. Photodynamic therapy—an up-to-date review. *Appl Sci*. 2021;11(8):3626. doi: [10.3390/app11083626](https://doi.org/10.3390/app11083626).
- Yu Z, Zhu M, Shu D, Zhang R, Xiang Z, Jiang A, et al. LncRNA PEG11 as aggravates cerebral ischemia/reperfusion injury after ischemic stroke through miR-342-5p/PFN1 axis. *Life Sci*. 2023;313:121276. doi: [10.1016/j.lfs.2022.121276](https://doi.org/10.1016/j.lfs.2022.121276).
- Correia JH, Rodrigues JA, Pimenta S, Dong T, Yang Z. Photodynamic therapy review: principles, photosensitizers, applications, and future directions. *Pharmaceutics*. 2021;13(9):1332. doi: [10.3390/pharmaceutics13091332](https://doi.org/10.3390/pharmaceutics13091332).
- Garg SJ, Hadziahmetovic M. Verteporfin photodynamic therapy for the treatment of chorioretinal conditions: a narrative review. *Clin Ophthalmol*. 2024;18:1701-16. doi: [10.2147/oph.S464371](https://doi.org/10.2147/oph.S464371).
- Jeong SB, Das R, Kim DH, Lee S, Oh HI, Jo S, et al. Anticancer effect of verteporfin on non-small cell lung cancer via downregulation of ANO1. *Biomed Pharmacother*. 2022;153:113373. doi: [10.1016/j.biopha.2022.113373](https://doi.org/10.1016/j.biopha.2022.113373).
- Kang MH, Jeong GS, Smoot DT, Ashktorab H, Hwang CM, Kim BS, et al. Verteporfin inhibits gastric cancer cell growth by suppressing adhesion molecule FAT1. *Oncotarget*. 2017;8(58):98887-97. doi: [10.18632/oncotarget.21946](https://doi.org/10.18632/oncotarget.21946).
- Song M, Zhu L, Zhang L, Ge X, Cao J, Teng Y, et al. Combination of molecule-targeted therapy and photodynamic therapy using nanoformulated verteporfin for effective uveal melanoma treatment. *Mol Pharm*. 2024;21(5):2340-50. doi: [10.1021/acs.molpharmaceut.3c01117](https://doi.org/10.1021/acs.molpharmaceut.3c01117).
- López MJ, Carbajal J, Alfaro AL, Saravia LG, Zanabria D, Araujo JM, et al. Characteristics of gastric cancer around the world. *Crit Rev Oncol Hematol*. 2023;181:103841. doi: [10.1016/j.critrevonc.2022.103841](https://doi.org/10.1016/j.critrevonc.2022.103841).
- Guan WL, He Y, Xu RH. Gastric cancer treatment: recent progress and future perspectives. *J Hematol Oncol*. 2023;16(1):57. doi: [10.1186/s13045-023-01451-3](https://doi.org/10.1186/s13045-023-01451-3).
- Chen B, Ma L, Paik H, Sirota M, Wei W, Chua MS, et al. Reversal of cancer gene expression correlates with drug efficacy and reveals therapeutic targets. *Nat Commun*. 2017;8:16022. doi: [10.1038/ncomms16022](https://doi.org/10.1038/ncomms16022).
- Hephzibah Cathryn R, Udhaya Kumar S, Younes S, Zayed H, George Priya Doss C. A review of bioinformatics tools and web servers in different microarray platforms used in cancer research. *Adv Protein Chem Struct Biol*. 2022;131:85-164. doi: [10.1016/bs.apcsb.2022.05.002](https://doi.org/10.1016/bs.apcsb.2022.05.002).
- Zhao Y, Ma T, Zou D. Identification of unique transcriptomic signatures and hub genes through RNA sequencing and integrated WGCNA and PPI network analysis in nonerosive reflux disease. *J Inflamm Res*. 2021;14:6143-56. doi: [10.2147/jir.S340452](https://doi.org/10.2147/jir.S340452).
- Rostami Nejad M, Bandarian F, Razi F, Razzaghi Z, Robati RM, Rezaei M, et al. Evaluation of laser intensity effect on photodynamic therapy efficacy. *J Lasers Med Sci*. 2024;15:e33. doi: [10.34172/jlms.2024.33](https://doi.org/10.34172/jlms.2024.33).
- Vafaei R, Arjmand B, Hamzeloo-Moghadam M, Rezaei Tavirani M, Razzaghi Z, Robati RM, et al. Assessment of photosensitizer concentration effects on the efficacy of photodynamic therapy. *J Lasers Med Sci*. 2023;14:e53. doi: [10.34172/jlms.2023.53](https://doi.org/10.34172/jlms.2023.53).

- 10.34172/jlms.2023.53.
15. Schmidt-Erfurth U, Hasan T. Mechanisms of action of photodynamic therapy with verteporfin for the treatment of age-related macular degeneration. *Surv Ophthalmol*. 2000;45(3):195-214. doi: [10.1016/s0039-6257\(00\)00158-2](https://doi.org/10.1016/s0039-6257(00)00158-2).
 16. Quinlan JA, Inglut CT, Srivastava P, Rahman I, Stabile J, Gaitan B, et al. Carrier-free, amorphous verteporfin nanodrug for enhanced photodynamic cancer therapy and brain drug delivery. *Adv Sci (Weinh)*. 2024;11(17):e2302872. doi: [10.1002/advs.202302872](https://doi.org/10.1002/advs.202302872).
 17. Strickler JH, Yoshino T, Graham RP, Siena S, Bekaii-Saab T. Diagnosis and treatment of ERBB2-positive metastatic colorectal cancer: a review. *JAMA Oncol*. 2022;8(5):760-9. doi: [10.1001/jamaoncol.2021.8196](https://doi.org/10.1001/jamaoncol.2021.8196).
 18. Yu D, Hung MC. Overexpression of ErbB2 in cancer and ErbB2-targeting strategies. *Oncogene*. 2000;19(53):6115-21. doi: [10.1038/sj.onc.1203972](https://doi.org/10.1038/sj.onc.1203972).
 19. Li X, Zhao L, Chen C, Nie J, Jiao B. Can EGFR be a therapeutic target in breast cancer? *Biochim Biophys Acta Rev Cancer*. 2022;1877(5):188789. doi: [10.1016/j.bbcan.2022.188789](https://doi.org/10.1016/j.bbcan.2022.188789).
 20. Ahmed MB, Alghamdi AAA, Islam SU, Lee JS, Lee YS. cAMP signaling in cancer: a PKA-CREB and EPAC-centric approach. *Cells*. 2022;11(13):2020. doi: [10.3390/cells11132020](https://doi.org/10.3390/cells11132020).
 21. Chen C, Yuan M, Xia L, Wu X, Zhong X, Zhang H, et al. Expression of crEbbp and Ep300 associated with tumor volume in patients with grade-3 glioma: a retrospective analysis. *Clin Med Insights Oncol*. 2024;18:11795549241287777. doi: [10.1177/11795549241287777](https://doi.org/10.1177/11795549241287777).
 22. LeBlanc F, Bennett J, Choi K, Starczynowski DT. Targeting CREB-binding protein (CREBBP) overcomes resistance to azacitidine and venetoclax therapy in acute myeloid leukemia (AML). *Blood*. 2023;142(Suppl 1):5765. doi: [10.1182/blood-2023-187063](https://doi.org/10.1182/blood-2023-187063).
 23. Mijanovic O, Petushkova AI, Brankovic A, Turk B, Solovieva AB, Nikitkina AI, et al. Cathepsin D-managing the delicate balance. *Pharmaceutics*. 2021;13(6):837. doi: [10.3390/pharmaceutics13060837](https://doi.org/10.3390/pharmaceutics13060837).
 24. Yang Y, Bagyinszky E, An SSA. Presenilin-1 (PSEN1) mutations: clinical phenotypes beyond Alzheimer's disease. *Int J Mol Sci*. 2023;24(9):8417. doi: [10.3390/ijms24098417](https://doi.org/10.3390/ijms24098417).