Journal of Carcinogenesis
Covering Genesis to Therapeutics

Journal of Carcinogenesis, ISSN: 1477-3163
2025; Vol 24: Issue 2s, https://carcinogenesis.com/

Therapeutic Potential of Withaferin A from Withania somnifera in Cancer: A Comprehensive Phytotherapeutic Review

Shivam Agarwal[1], Harshit Thakur[2], Rashmi[3], Baijnath Das[4], Praveen Katiyar[*5]

[1]Ph.D. Scholar, School of Health Sciences, Chhatrapati Shahu Ji Maharaj University, Kanpur, Uttar Pradesh, India, 208024

Email ID: shivamagarwal50283@gmail.com / ORCID ID: 0000-0002-7891-9389

[2] Assistant Professor, School of Allied health science, Galgotias university, Greater Noida, Gautam Buddh Nagar, Uttar Pradesh, India, 203201.

Email ID: harshitthakur736@gmail.com

[3] Professor, Physics Wallah IOI School of Healthcare, Email ID: rashmi23july@gmail.com

[4]Lecturer, College of Paramedical Sciences, Teerthanker Mahaveer University, Moradabad, Uttar Pradesh, India, 244001.

Email ID: baijnathdasbiochemaiims@gmail.com

[5]Associate Professor, School of Health Sciences, Chhatrapati Shahu Ji Maharaj University, Kanpur, Uttar Pradesh, India, 208024

ORCID: 0000-0001-5902-8073

***Corresponding Author:**

Praveen Katiyar

Email ID: drpraveenkatiyar@gmail.com

ABSTRACT

Withaferin A, a steroidal lactone and major withanolide of *Withania somnifera* (Ashwagandha), has gained considerable attention for its potent anticancer potential. Abundantly present in the leaves and roots, WA exhibits a broad pharmacological profile encompassing anti-inflammatory, immunomodulatory, antioxidant, and cytotoxic properties. Extensive preclinical studies reveal that WA exerts anticancer effects through multiple, interrelated mechanisms. These include apoptosis induction via activation of p53, Bax, and caspases while inhibiting Bcl-2; cell cycle arrest at the G2/M phase through downregulation of cyclins and cyclin-dependent kinases; oxidative stress modulation by enhancing reactive oxygen species generation and impairing antioxidant defenses; oncogenic signaling suppression involving inhibition of NF-κB, STAT3, Akt, and Notch pathways; and anti-metastatic/anti-angiogenic activity through downregulation of matrix etalloproteinase-9 and vascular endothelial growth factor. Notably, WA demonstrates selective cytotoxicity toward cancer cells while sparing normal tissues, making it an attractive candidate for cancer chemoprevention and therapy. However, clinical translation remains limited due to bioavailability challenges, warranting advanced formulation strategies and well-designed clinical trials. This review provides a comprehensive analysis of WA's phytochemistry, molecular mechanisms, and therapeutic relevance, underscoring its potential as a multi-targeted phytotherapeutic agent in oncology.

Keywords: *Withaferin A, Withania somnifera, apoptosis, cell cycle arrest, oxidative stress, NF-κB inhibition, anti-angiogenesis*

How to Cite: Anusha PK, (2025) Therapeutic Potential of Withaferin A from Withania somnifera in Cancer: A Comprehensive Phytotherapeutic Review, *Journal of Carcinogenesis*, *Vol.24, No.2s*, 640-646

1. INTRODUCTION

Ashwagandha holds a prominent position in Ayurveda and is revered as one of the most potent rejuvenating herbs (1). The name is derived from Sanskrit—'Ashwa' meaning horse and 'Gandha' meaning smell—referring to the distinctive odor of the root and its traditional belief of imparting the strength and vitality of a stallion (2). *Withania somnifera*, commonly known as Ashwagandha, is a short, woody shrub that typically grows to a height of 35–75 cm. The plant exhibits radial branching from a central stem and has ovate, dull green leaves measuring approximately 10–12 cm in length (see figure 1

(a)). Its flowers are small, greenish-yellow in color, and round in shape, typically occurring in clusters (axillary cymes). Each flower measures about 4–6 mm in diameter. The fruits are smooth, red berries enclosed within an enlarged green calyx (see figure 1 (b)). The roots are stout, long, tuberous, fleshy, and whitish brown with a distinct horse-like aroma, which is a characteristic feature of the plant see Figure 1 (3).

Figure 1. *Withania somnifera* in natural habitat (a) and fruits (b) (Source; Srivastava et al. 2018) (4)

2. WITHAFERIN A

Withaferin A (WA) is a naturally occurring steroidal lactone classified within the withanolide group of bioactive compounds (5). It was first isolated in 1962 from the leaves of *Withania somnifera* (Ashwagandha or Indian winter cherry), a medicinal plant belonging to the Solanaceae family and extensively used in traditional Ayurvedic medicine for centuries to manage inflammation, stress, aging, and immune-related disorders (6). Among the more than 40 withanolides identified in *W. somnifera*, WA is considered the most abundant and pharmacologically active constituent, exhibiting a broad spectrum of therapeutic effects, including anti-inflammatory, immunomodulatory, and anticarcinogenic properties (7). The anticancer potential of WA was first reported in the early 1970s using the Ehrlich ascites tumor model, and subsequent preclinical studies have consistently demonstrated its potent cytotoxicity against diverse cancer cell types, accompanied by minimal toxicity toward normal cells (8). This selective cytotoxicity, coupled with its ability to modulate multiple molecular targets and signaling pathways, positions WA as a promising candidate for both cancer chemoprevention and therapy (9).

3. WITHANIA SOMNIFERA PLANT

3.1 Habitat and Distribution

Withania somnifera is widely distributed across the Indian subcontinent and is found in various agro-climatic regions ranging from northern to southern India. It is also cultivated extensively for medicinal and commercial purposes, particularly for use in traditional and modern herbal formulations. Beyond India, the plant is naturally found in regions of Nepal, China, and Yemen, where it thrives in dry and subtropical climates (10).

3.2 Taxonomic Classification

Botanically, *Withania somnifera* is classified as follows:

Kingdom: Plantae,

Subkingdom: Tracheobionta ,

Super division: Spermatophyta,

Division: Magnoliophyta,

Class: Magnoliopsida,

Subclass: Rosidae,

Order: Solanales,

Family: Solanaceae,

Genus: *Withania,*

Species: *Withania somnifera* (11)

3.3 Life Cycle:

Withania somnifera is a short-day, perennial plant commonly grown as an annual for commercial purposes. The life cycle consists of:

- **Germination:** Occurs 7–10 days after sowing.
- **Vegetative phase:** Lasts for 30–60 days.
- **Flowering phase:** Begins 60–70 days after sowing.
- **Fruiting and maturity:** Harvested around 150–180 days post-sowing, when berries turn red and the roots are fully developed (11).

3.4 Biological Function

The biological functions of Withania somnifera are diverse and therapeutically significant, making it a vital herb in traditional and modern medicine. It exhibits adaptogenic, anti-inflammatory, antioxidant, and neuroprotective activities, contributing to its role in stress reduction and cognitive enhancement. Additionally, the plant has demonstrated antitumor, anti-ageing, hepatoprotective, and antimicrobial effects, supporting its utility in cancer, liver disorders, and infectious diseases. Withania somnifera also promotes macrophage activation and musculotropic activity, beneficial in immunity and muscular health. These broad-spectrum biological actions are visually represented in Figure 2, which highlights the multifaceted therapeutic potential of the plant (12).

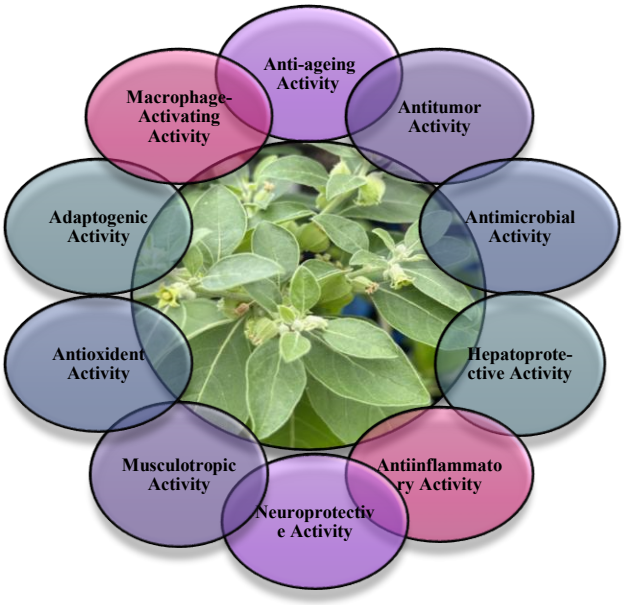

Figure 2. *Biological functions of Withania somnifera.* The diagram illustrates the major pharmacological activities of *Withania somnifera*, including anti-ageing, antitumor, antimicrobial, hepatoprotective, anti-inflammatory, neuroprotective, musculotropic, antioxidant, adaptogenic, and macrophage-activating activities. These properties contribute to the herb's broad therapeutic potential in managing oxidative stress, inflammation, cancer, neurodegenerative disorders, immune modulation, and general health enhancement.

4. PHYTOCHEMICAL PROFILE WITHANIA SOMNIFERA FOR WITHAFERIN A

Withania somnifera produces a diverse spectrum of bioactive metabolites, notably withanolides, withanosides, alkaloids, steroidal lactones, phenolics, fatty acids, and sterols, each contributing to its pharmacological potential (13). These compounds are unevenly distributed among different plant parts, with WA being particularly concentrated in the leaves and, to a lesser extent, in the roots (14).

4.1 Distribution in Plant Parts

- Leaves — Richest source of WA, also containing withanolide derivatives (e.g., withanolide A, B, C) and withanosides with strong anti-inflammatory and anticancer activities.

- Roots — Contain WA along with numerous withanolides, withanosides, and alkaloids that contribute to adaptogenic and anticancer effects.
- Fruits — Contain withanolides, withanamides, fatty acids, and sterols, contributing to antioxidant and neuroprotective functions, but relatively low WA content.
- Stem bark — Contains unique withanolide analogues, though WA presence is minimal.

4.2 Withaferin A in the Phytochemical Context

Among the more than 40 withanolides identified in *W. somnifera*, WA stands out as:

- The most abundant withanolide in leaves
- One of the most pharmacologically potent constituents
- A multifunctional therapeutic molecule with activities ranging from anti-inflammatory to antitumor

Its relatively high natural abundance in aerial parts makes WA more accessible for extraction compared to other rare withanolides, enhancing its potential for pharmaceutical development (13).

Chemical Structure:

Withaferin A has a complex steroidal structure with a molecular formula of C28H38O6 and a molecular weight of about 470.6 g/mol (Figure 3). Its structure includes four cycloalkane rings (three cyclohexane rings and one cyclopentane ring), typical of steroid frameworks (15). Key reactive features contributing to its biological activity include:

- An unsaturated ketone-containing A ring with a double bond, which is highly reactive.
- An epoxide group in the B ring, which also contributes to its cytotoxicity.
- An unsaturated delta-lactone ring involving oxidation at carbons 22 and 26.
- Hydroxyl groups at specific positions (4 and 27), which modulate its solubility and activity.

These structural features confer Withaferin A its bioactivity, particularly its ability to interact with cellular proteins and disrupt cancer cell functions (16).

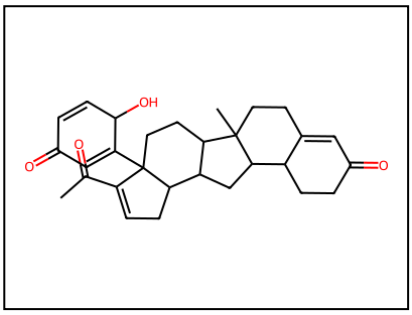

Figure 3. Chemical Structure of Withaferin A.

Phytotherapeutic Properties:

As a bioactive molecule from Withania somnifera, Withaferin A exhibits several medicinal properties relevant to cancer therapy, including:

- Anti-inflammatory activity by inhibiting key inflammatory pathways.
- Immunomodulatory effects that enhance the body's ability to combat tumors.
- Antioxidant properties that help in maintaining cellular redox balance.
- Antiproliferative and pro-apoptotic activities that suppress tumor growth (17).

Mechanisms of Action Against Cancer of Withaferin A

Withaferin A, a bioactive steroidal lactone isolated from *Withania somnifera*, exhibits potent anticancer activity through multiple, interrelated molecular mechanisms targeting key cellular pathways involved in tumor initiation, progression, and metastasis (18) (Figure 4).

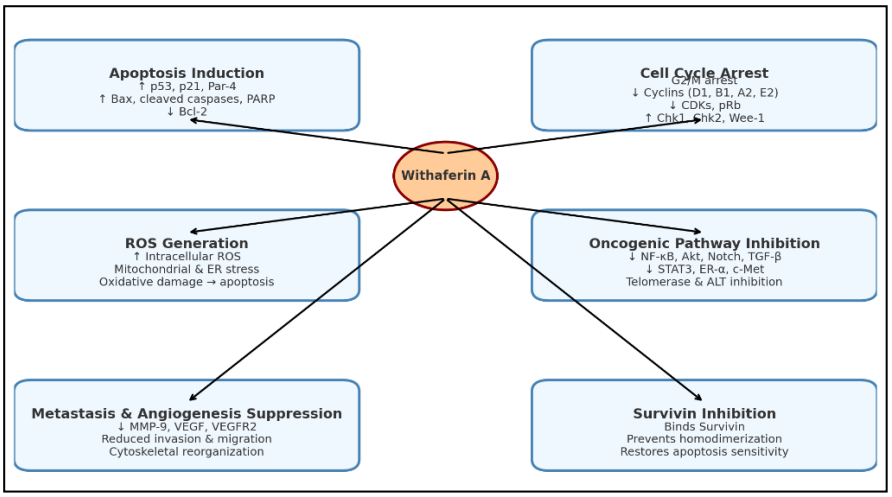

Figure 3. Mechanisms of Action Against Cancer of Withaferin A.

A major mechanism involves the induction of apoptosis via both intrinsic and extrinsic pathways, characterized by the activation of pro-apoptotic proteins such as Bax, cleaved caspase-3, -8, and -9, and PARP, along with the tumor suppressors p53, p21, and prostate apoptosis response-4 (Par-4), (19)while concomitantly suppressing anti-apoptotic proteins like Bcl-2 (20). In parallel, WA induces cell cycle arrest predominantly at the G2/M phase by downregulating cyclins (D1, B1, A2, E2) and cyclin-dependent kinases (CDK1, Cdc2), inhibiting phosphorylation of the retinoblastoma protein (pRb), and activating checkpoint kinases (Chk1, Chk2) and Wee-1 (21). Additionally, it disrupts microtubule dynamics through direct binding to β-tubulin and causes reorganization of cytoskeletal proteins such as vimentin and annexin-A2, ultimately leading to mitotic catastrophe (22).

Another crucial mechanism is the generation of excessive intracellular reactive oxygen species (ROS), which induces oxidative stress, mitochondrial and endoplasmic reticulum dysfunction, and macromolecular damage, culminating in apoptosis, ferroptosis, or paraptosis (23,24). WA also impairs antioxidant defense mechanisms by modulating the Keap1–Nrf2 axis, thereby selectively sensitizing cancer cells to oxidative injury (25). Furthermore, WA exerts broad inhibitory effects on multiple oncogenic signaling cascades, including suppression of the NF-κB pathway (inhibiting p65 nuclear translocation) (26), Akt signaling (reducing proliferation, migration, and epithelial-to-mesenchymal transition), Notch and TGF-β pathways (limiting tumor progression and invasion), and other regulators such as STAT3, estrogen receptor-α, and c-Met (27). Notably, WA also targets telomere maintenance mechanisms in both telomerase-positive and ALT-dependent cancer cells by disrupting ALT-associated promyelocytic leukemia nuclear bodies and downregulating the MRN complex protein NBS-1 (28).

Inhibition of metastasis and angiogenesis represents another important facet of WA's anticancer profile (29). By downregulating matrix metalloproteinase-9 (MMP-9), WA restricts extracellular matrix degradation, thereby limiting invasion, while suppression of vascular endothelial growth factor (VEGF) and VEGFR2 signaling impedes angiogenesis and tumor vascularization (30). Moreover, WA directly interacts with Survivin, an inhibitor of apoptosis protein frequently overexpressed in cancers, preventing its homodimerization and restoring apoptotic sensitivity in resistant tumor cells (31). Importantly, WA demonstrates preferential cytotoxicity toward malignant cells while sparing normal cells, likely due to its selective targeting of dysregulated oxidative stress responses and aberrant signaling networks unique to cancer cells (32). This multimodal mechanism of action positions WA as a promising candidate for development into a broad-spectrum anticancer therapeutic (16).

Conclusion

Withaferin A, the most abundant and pharmacologically potent withanolide of *Withania somnifera*, exhibits a remarkable spectrum of anticancer activities through multi-targeted mechanisms, including induction of apoptosis, cell cycle arrest, cytoskeletal disruption, oxidative stress modulation, and inhibition of metastasis and angiogenesis. Its preferential cytotoxicity toward malignant cells, coupled with low toxicity to normal tissues, underscores its potential as a safe and effective phytotherapeutic agent. The rich natural abundance of Withaferin A in the aerial parts of *W. somnifera*, particularly leaves, facilitates sustainable sourcing for pharmaceutical development. Despite compelling preclinical evidence, translation into clinical oncology remains limited, with challenges such as bioavailability, pharmacokinetics, and formulation stability requiring further optimization. Future research should focus on well-designed clinical trials, nanoformulation strategies, and combinatorial regimens with existing chemotherapeutics to fully harness Withaferin A's therapeutic promise in cancer prevention and treatment.

REFERENCES

[1] Mikulska P, Malinowska M, Ignacyk M, Szustowski P, Nowak J, Pesta K, et al. Ashwagandha (Withania somnifera)—Current Research on the Health-Promoting Activities: A Narrative Review. Pharmaceutics. 2023 Mar 24;15(4):1057.

[2] Paul S, Chakraborty S, Anand U, Dey S, Nandy S, Ghorai M, et al. Withania somnifera (L.) Dunal (Ashwagandha): A comprehensive review on ethnopharmacology, pharmacotherapeutics, biomedicinal and toxicological aspects. Biomedicine & Pharmacotherapy. 2021 Nov 1;143:112175.

[3] A GA, D FL, Dp A, A MV, Ci FL. Effects of Withania somnifera (Ashwagandha) on Hematological and Biochemical Markers, Hormonal Behavior, and Oxidant Response in Healthy Adults: A Systematic Review. Current nutrition reports [Internet]. 2023 Sep [cited 2025 Aug 14];12(3). Available from: https://pubmed.ncbi.nlm.nih.gov/37428341/

[4] Srivastava M, Gupta S, Dixit S, Yadav N, Yadav V, Singh H, et al. Withania somnifera (Ashwagandha): A wonder herb with multiple medicinal properties. Asian Journal of Pharmacy and Pharmacology. 2018 Apr 1;4:123–30.

[5] CSIR-Central Institute of Medicinal and Aromatic Plants, Lucknow-226015, India, Shukla A, Srivastava S, Dev K. Review on chemistry, pharmacology, toxicity and future prospectives of withaferin A. JMAPS. 2023 Sep 5;45(3):108–22.

[6] Umadevi M, Rajeswari R, Rahale CS, Selvavenkadesh S, Pushpa R, Kumar KPS. Traditional And Medicinal Uses of Withania Somnifera. 2012;1(9).

[7] IJCSP25A1144.pdf [Internet]. [cited 2025 Aug 14]. Available from: https://www.rjpn.org/ijcspub/papers/IJCSP25A1144.pdf

[8] Hahm ER, Kim SH, Singh KB, Singh K, Singh SV. A Comprehensive Review and Perspective on Anticancer Mechanisms of Withaferin A in Breast Cancer. Cancer Prev Res (Phila). 2020 Sep;13(9):721–34.

[9] Hashem S, Ali TA, Akhtar S, Nisar S, Sageena G, Ali S, et al. Targeting cancer signaling pathways by natural products: Exploring promising anti-cancer agents. Biomedicine & Pharmacotherapy. 2022 Jun 1;150:113054.

[10] Kumari - Medicinal Properties and Seed Germination Behaviou.pdf [Internet]. [cited 2025 Aug 14]. Available from: https://biothink.in/wp-content/uploads/2025/01/biothink-2025-02-updated.pdf

[11] wikipedia. Withania somnifera. In: Wikipedia [Internet]. 2025 [cited 2025 Aug 14]. Available from: https://en.wikipedia.org/w/index.php?title=Withania_somnifera&oldid=1304092900

[12] Nair AR, Praveen N. Biochemical and phytochemical variations during the growth phase of Withania somnifera (L.) Dunal.

[13] Saleem S, Muhammad G, Hussain MA, Altaf M, Bukhari SNA. Withania somnifera L.: Insights into the phytochemical profile, therapeutic potential, clinical trials, and future prospective. Iran J Basic Med Sci. 2020 Dec;23(12):1501–26.

[14] Zagoskina NV, Zubova MY, Nechaeva TL, Kazantseva VV, Goncharuk EA, Katanskaya VM, et al. Polyphenols in Plants: Structure, Biosynthesis, Abiotic Stress Regulation, and Practical Applications (Review). Int J Mol Sci. 2023 Sep 9;24(18):13874.

[15] Kumar S, Mathew SO, Aharwal RP, Tulli HS, Mohan CD, Sethi G, et al. Withaferin A: A Pleiotropic Anticancer Agent from the Indian Medicinal Plant Withania somnifera (L.) Dunal. Pharmaceuticals (Basel). 2023 Jan 22;16(2):160.

[16] Sultana T, Okla MK, Ahmed M, Akhtar N, Al-Hashimi A, Abdelgawad H, et al. Withaferin A: From Ancient Remedy to Potential Drug Candidate. Molecules. 2021 Dec 20;26(24):7696.

[17] Atteeq M. Evaluating anticancer properties of Withaferin A-a potent phytochemical. Front Pharmacol. 2022;13:975320.

[18] Kamel EM, Ali MAM, Allam AA, Ahmed NA, Aba Alkhayl FF, Lamsabhi AM. Disrupting the Hsp90-Cdc37 axis: a selective strategy for targeting oncogenic kinases in cancer. RSC Adv. 2025 Jun 4;15(24):19376–91.

[19] Capuozzo M, Santorsola M, Bocchetti M, Perri F, Cascella M, Granata V, et al. p53: From Fundamental Biology to Clinical Applications in Cancer. Biology. 2022 Sep 6;11:1325.

[20] Mosadegh M, Noori Goodarzi N, Erfani Y. A Comprehensive Insight into Apoptosis: Molecular Mechanisms, Signaling Pathways, and Modulating Therapeutics. Cancer Invest. 2025 Jan;43(1):33–58.

[21] Lee D, Hokinson D, Park S, Elvira R, Kusuma F, Lee JM, et al. ER Stress Induces Cell Cycle Arrest at the G2/M Phase Through eIF2α Phosphorylation and GADD45α. Int J Mol Sci. 2019 Dec 13;20(24):6309.

[22] Stanton RA, Gernert KM, Nettles JH, Aneja R. Drugs That Target Dynamic Microtubules: A New Molecular

Perspective. Med Res Rev. 2011 May;31(3):443–81.

[23] Hong Y, Boiti A, Vallone D, Foulkes NS. Reactive Oxygen Species Signaling and Oxidative Stress: Transcriptional Regulation and Evolution. Antioxidants (Basel). 2024 Mar 1;13(3):312.

[24] Zorov DB, Juhaszova M, Sollott SJ. Mitochondrial reactive oxygen species (ROS) and ROS-induced ROS release. Physiol Rev. 2014 Jul;94(3):909–50.

[25] Tu W, Wang H, Li S, Liu Q, Sha H. The Anti-Inflammatory and Anti-Oxidant Mechanisms of the Keap1/Nrf2/ARE Signaling Pathway in Chronic Diseases. Aging Dis. 2019 Jun 1;10(3):637–51.

[26] Xing Z, Su A, Mi L, Zhang Y, He T, Qiu Y, et al. Withaferin A: A Dietary Supplement with Promising Potential as an Anti-Tumor Therapeutic for Cancer Treatment - Pharmacology and Mechanisms. Drug Des Devel Ther. 2023 Sep 21;17:2909–29.

[27] Maharati A, Moghbeli M. PI3K/AKT signaling pathway as a critical regulator of epithelial-mesenchymal transition in colorectal tumor cells. Cell Commun Signal. 2023 Aug 14;21:201.

[28] Recagni M, Bidzinska J, Zaffaroni N, Folini M. The Role of Alternative Lengthening of Telomeres Mechanism in Cancer: Translational and Therapeutic Implications. Cancers (Basel). 2020 Apr 11;12(4):949.

[29] El-Kenawi AE, El-Remessy AB. Angiogenesis inhibitors in cancer therapy: mechanistic perspective on classification and treatment rationales. British Journal of Pharmacology. 2013 Oct 1;170(4):712.

[30] Huang H. Matrix Metalloproteinase-9 (MMP-9) as a Cancer Biomarker and MMP-9 Biosensors: Recent Advances. Sensors (Basel). 2018 Sep 27;18(10):3249.

[31] Albadari N, Li W. Survivin Small Molecules Inhibitors: Recent Advances and Challenges. Molecules. 2023 Feb 1;28(3):1376.

[32] Blagosklonny MV. Selective protection of normal cells from chemotherapy, while killing drug-resistant cancer cells. Oncotarget. 2023 Mar 11;14:193–206.