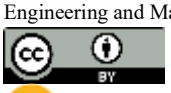
Anticancer Properties of Thevetia

Sristy , Dr . Manesh singh
department of Pharmacology

How to Cite this Article:
Sristy, (2026). Anticancer Properties of Thevetia. International Journal of Creative and Open Research in Engineering and Management, 02(05). https://doi.org/10.55041/ijcope.v2i5.759



https://doi.org/ 10.55041/ijcope.v2i5.759

Abstract

Thevetia peruviana (syn. Cascabela thevetia), a plant belonging to the Apocynaceae family, has long been recognized for its dual nature as both a potent poison and a source of traditional medicines. In recent years, scientific interest has shifted towards its significant potential in oncology. This review synthesizes the current body of research on the anticancer properties of T. peruviana. The plant is a rich reservoir of bioactive compounds, most notably cardiac glycosides (cardenolides) such as thevetin, peruvoside, and neriifolin, which are concentrated in the seeds and other plant parts. In addition, various extracts contain significant amounts of phenolic compounds, flavonoids, and other secondary metabolites. Numerous in vitro studies have demonstrated that extracts from the fruit, leaves, seeds, and latex of T. peruviana exhibit potent cytotoxic and antiproliferative activity against a wide spectrum of human cancer cell lines, including those of the breast, colon, lung, prostate, and liver. The primary mechanism of action is attributed to the inhibition of the Na+/K+-ATPase pump by cardiac glycosides, which disrupts cellular ion homeostasis and triggers downstream signaling cascades. These events culminate in the induction of apoptosis, characterized by caspase activation and PARP cleavage, as well as cell cycle arrest, typically at the G2/M phase. This review consolidates the phytochemical profile of T. peruviana, details its cytotoxic efficacy with specific IC50 values, and elucidates the molecular mechanisms underlying its anticancer effects, highlighting its promise as a source for the development of novel chemotherapeutic agents.

1. INTRODUCTION

Thevetia peruviana (yellow oleander) is a tropical decorative plant (family Apocynaceae), which contains highly cardiotoxic glycosids. Although toxic T. peruviana was given medicinal importance in traditional practices but was used to treat skin conditions and dissolve tumors. Recent studies within the last decade have revealed that T. peruviana extracts and components have a great potential in the context of anticancer. More specifically, T. peruviana contains bioactive compounds - in particular, cardiac glycosides (cardenolides) such as peruvoside, thevetin, neriifolin, etc. - which, in addition to affecting important molecular pathways, have a cytotoxic effect on cancer cells. The review summarises the results of in vitro cell culture, in vivo animal model and any developing clinical knowledge in the anticancer activity of T. peruviana. Mechanistic evidences that we are interested in include induction of apoptosis (programmed cell death), modulation of oxidative stress, arrest of the cell cycle, and other cellular changes which are based on its antineoplastic effects.

2. In Vitro Anticancer Activity

Various researchers have shown that T. peruviana crude extracts cause cytotoxicity in various human cancer cells. As an example, a methanolic extract of T. peruviana fruit exhibited high growth inhibition in human prostate cells, (PC-3), breast cells, (MCF-7), colon and lung cancer cells with 50 percent inhibitory concentration (IC 50) within the low range of microgram-per-milliliter. According to the standards of U.S. NCI, these IC50 (less than 20 ug/mL) make the crude extract highly active. It is interesting to note that Ramos-Silva et al. (2017) found the IC50 of prostate cancer cells to be 1.9 ug/mL, breast, colon, and lung carcinoma cells to be around 5-12 ug/mL, but the extract was significantly less toxic to normal fibroblasts (IC50 >1,000 ug/mL). This selectivity indicates that T. peruviana extract has the ability to select tumor cells and avoid normal cells. In line with this, it was found that the colony formation (clonogenicity) in treated cancer cells reduced by about 80 percent compared to the controls, but healthy cells were not affected. T. peruviana fruit extract was also found to significantly prevent the migration of cancer cells in wound- healing experiments, but did not disrupt the normal cell movement. These results depict a wide anti- proliferative and anti-migratory action of the fruit extract in vitro.

The same effects are created by other parts of plants. Recent research on T. peruviana leaves found there to be lots of polyphenols (e.g. gallic acid, chlorogenic acid, quercetin, rutin) present and has found the leaf extract to be significantly cytotoxic against colon carcinoma cells (HCT-116) (IC50 of the leaf extract of about 39 ug/mL). The activity was less pronounced against lung (A-549) and breast (MCF-7) cells (IC50 - 110 ug/mL). At the same time, T. peruvina fruits were found to contain high levels of flavonoid (rutin, quercetin, naringin, etc.) and phenolic acid (latex). Latex exhibited strong antioxidant (DPPH radical scavenging IC50 = +43.9 ug/mL), and significant in vitro cytotoxicity: fresh latex (1 mg/mL) was able to induce cell death in PC-3 prostate and MCF-7 breast cancer cells (approximately 97 and 96 percent respectively) and had an IC50 of approximately 40-48 ug/mL. Microscopic analysis had shown that exposure to latex resulted in significant morphological modifications (cytotoxic concentrations) in the cancer cells (cell shrinkage, loss of adhesion). These data (Al-Rajhi et al., 2022) prove the idea that T. peruviana latex has bioactive compounds and kills cancer cells in vitro, which is probably helped by its antioxidant phytochemicals that may regulate the redox status of the cell.

Even stronger effects of anticancer activity have been demonstrated by purified T. peruvana cardiac glycosides seeds. Cheng et al. (2016) extracted a few cardenolides in T. peruviana seeds, and one of them (a novel compound named as compound 1) presented sub-micromolar IC50 values (0.05-0.15 uM) against human lung, gastric and pancreatic cancer cells. The compound did not only suppress the proliferation of cancer cells at nanomolar doses, but also it highly caused the apoptosis of the treated cells. Interestingly, it specifically was more toxic to cancer cells than normal hepatocytes. Other glycoside peruvoside T. peruvina that is known has been studied in other types of cancer cells. In vitro antiproliferative effect of peruvoside was observed in human breast, lung and liver carcinoma cells via loss of viability and cell cycle arrest and this was demonstrated to be dose dependent. Indicatively, peruvoside had low nanomolar potency (IC50 of peruvoside is about 9 nM at 72 h) in the context of MCF-7 breast cancer cells and a marked reduction in clonogenic cell survival. Table 1 is a summary of some in vitro studies on T. peruviana. All these cumulatively prove that extracts or compounds derived by T. peruviana have strong cytotoxic activity on cancer cells and that their mechanisms need a more thorough review.

Table 1: Selected In Vitro Anticancer Studies of *T. peruviana*

T. peruviana Source (Study)	Cancer Cell Models	Key Findings (IC_{50}, Effects)	Reference
Fruit methano extract (Ramos-Silva 2017)	Prostate (PC-3), Breast (MCF-7), Colon, Lung carcinoma Normal fibroblasts, Vero cells	IC_{50}: 1.9 µg/mL (prostate), 5– 12 µg/mL (others). Potent cytotoxicity in all cancer lines (<20 µg/mL) with negligible effect on normal cells. Induced >70–80% reduction in cancer colony formation and migration. Triggered apoptotic DNA fragmentation in cancer cells.	Ramos- Silva et al., 2017
Leaf extract (El Gizawy 2023)	Colon (HCT-116), Lung (A-549), Breast (MCF-7) cells	IC_{50}: 39.3 µg/mL (HCT-116); ~93– 110 µg/mL (A-549, MCF-7). Contains high phenolic content (gallic, chlorogenic acids) and flavonoids (quercetin, rutin). Moderately selective for cancer vs. normal cells (not reported toxic to normals).	El Gizawy et al., 2023
Latex (milky sap) (Al-Rajhi 2022)	Prostate (PC-3), Breast (MCF-7) cells *(+ various microbes)*	IC_{50}: 48.3 µg/mL (PC-3), 40.3 µg/mL (MCF-7). ~96–97% cell death at 1000 µg/mL. Rich in flavonoids	Al-Rajhi et al., 2022
T. peruviana Source (Study)	Cancer Cell Models	Key Findings (IC_{50}, Effects)	Reference
		(rutin, quercetin) and phenolics. Displays antioxidant activity (DPPH IC_{50} ~43.9 µg/mL). Causes cell shrinkage and loss of adhesion at cytotoxic doses.	
Seed cardiac glycoside "compound 1" (Cheng 2016)	Lung (P-15), Gastric (MGC-803), Pancreatic (SW1990) cancer cells Normal LO2 hepatocytes	IC_{50}: 0.05–0.15 µM (50–150 nM) in cancers. Selectively inhibits cancer cell proliferation with minimal effect on normal cells. Induces dose- dependent apoptosis in MGC-803 gastric cells.	Cheng et al., 2016

Flower ethanolic extract (Managit 2017)	Cervical (HeLa) cells (with TNF-α or TRAIL co-treatment)	Alone, modest cytotoxicity; but **sensitizes** cells to TNF-α and TRAIL-induced apoptosis. Combined treatment triggers extensive caspase activation and BID cleavage. Downregulates anti-apoptotic proteins (Mcl-1, Bcl-x_L XIAP, survivin) with TNF-α, and Bcl-x_L with TRAIL, thereby overcoming death ligand resistance.	Managit et al., 2017
Peruvoside (cardenolide) – isolated (Feng 2016; Reddy 2020)	Leukemia (KG1a, K562); Breast (MCF-7); Lung (A549); Liver (HepG2) cells Normal blood lymphocytes	Low-nanomolar antiproliferative activity in solid tumors (IC_{50} ~5– 50 nM) and leukemia (KG1a IC_{50} ~26 nM). Causes **cell cycle arrest** (G_0/G_1 in MCF-7; G_2/M in leukemia) and DNA damage. Induces apoptosis via caspase-8, -9, and -3 activation and PARP cleavage. Modulates multiple signaling pathways: inhibits Wnt/β-catenin (↓cyclin D1, c-Myc),	Feng et al. 2016; Reddy et al., 2020

T. peruviana Source (Study)	Cancer Cell Models	Key Findings (IC_{50}, Effects)	Reference
		blocks PI3K/AKT/mTOR (autophagy) and MEK/ERK pathways, and downregulates NF-κB and JAK-STAT signaling. Exhibits minimal cytotoxicity on normal lymphocytes, suggesting a therapeutic window.	

3. Mechanistic Insights

The anticancer action of T. peruviana is supported by several molecular mechanisms, the main ones being the induction of the process of apoptosis and disruption of the cancer cell survival pathways. One of the common motifs of the studies is apoptosis induction. The T. peruviana extract treatment results in classic apoptotic features in cancerous cells such as blebbing of the membrane, condensing of the chromatin, and fragmentation of the DNA. Ramos-Silva et al. also reported an apparent DNA laddering in agarose gels of extract treated cancer cells, similar to that of apoptotic DNA fragmentation in doxorubicin and untreated cells exhibited intact DNA. Dual staining of fruit extract with acridine orange and ethidium bromide was used to confirm that the fruit extract has a rapid effect of increasing the percentage of cells in early apoptosis (by an average of 40% in 4 h) in lung and prostate cancer cultures. T. peruviana is a mechanistic trigger of intrinsic (mitochondrial) and extrinsic (death receptor) apoptosis. Cheng et al. (2016) showed that the seed- derived glycoside activates the caspase-9 and caspase-3, whilst increasing pro-apoptotic Bax/Bcl-2 ratio, to gastric carcinoma cells. This implies involvement of the intrinsic apoptotic pathway that ends up releasing cytochrome c in the mitochondria and activation of caspase cascade. At the same time, that compound induced G2/M cell-cycle arrest (discussed below) which tends to enhance apoptotic signaling. In agreement, Feng et al. (2016) have demonstrated that peruvoside induces caspase-8 activation and Poly(ADP-ribose) polymerase (PARP) cleavage in leukemia cells, which implicates extrinsic apoptosis through death receptors. In fact, T. peruviana flower extract was identified to sensitize HeLa cervical cancer cells to extrinsic apoptosis receptor-TRAIL -ligand TNF-a, and TRAIL -ligand following downregulation of major anti-apoptotic proteins (Mcl-1, Bcl-xL, XIAP, survivin) which provide resistance. The treatment of the extract + TNF or TRAIL resulted in a strong caspase cascade and BID (a BH3-only protein connected with extrinsic and intrinsic pathways) activation, and the death of cancer cells was significantly increased.

All these data support the hypothesis that T. peruviana compounds have the potential to reduce the apoptotic threshold of cancer cells, which promotes programmed cell death via a variety of overlapping pathways.

4. Cell Cycle Arrest

The other process through which T. peruviana elicits antiprolliferative actions is through impairing the cell cycle to suppress the division of cancer cells. Various research findings provide information on the cell cycle blockage at different stages relative to the type of compound and cell environment. Peruvoside and cardenolides are widely used and usually result in a G0/G1 phase blockage when used on solid tumor cells. Convallatoxin (CT) and peruvoside (PS) treatment resulted in a more than 80% increase in the percentage of the cells in G0/G1, and decreases in S and G2/M cells, respectively in hormone responsive MCF-7 breast cancer cells. Western blot validations showed that both compounds suppressed cyclin D1 and CDK4 - major contributors of G1-S transition - in accordance with a G1 arrest. Figure 1 depicts the dose and time-related viability reduction in MCF-7 cells with the application of CT and PS, the effects of the two on colony formation, and cell-cycle. CT and PS also induced G0/G1 arrest in triple-negative models of breast cancer and had better efficacy in highly aggressive MDA-MB-468 (African American origin) over MDA-MB-231 (Caucasian origin), indicating some cell-line-dependent sensitivities. On the flip side, G2/M arrest can be induced in some cancers by some T. peruviana compounds. The seed glycoside of Cheng induced G2/M arrest in MGC-803 gastric cancer cells, and Feng et al. found that peruvoside elevated the G2/M population of acute myeloid leukemia cells (KG1a) and induced the cocktail of CDKN1A (p21Cip1) cyclin-dependent kinase blocking. This branch implies that T. peruviana cardenolides may interfere with the cell cycle events at various stages, which may be cancer-specific. These agents reduce proliferation of cancer cells by arresting G0/G1 or G2/M cells and can induce apoptosis (since prolonged arrest may cause apoptosis).

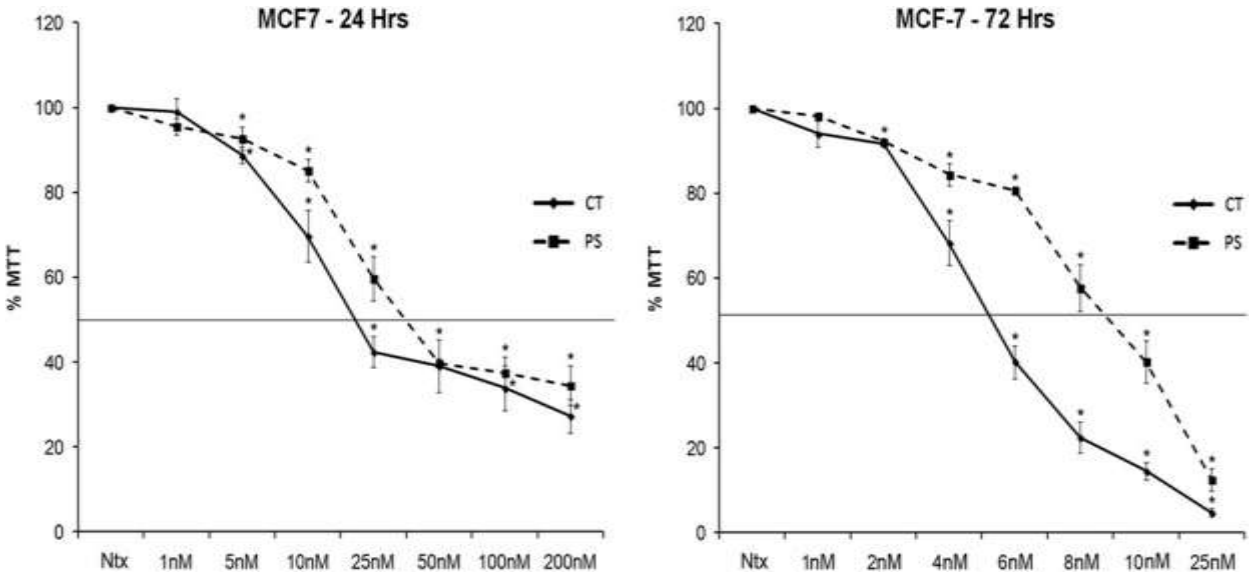


Figure 1: *Dose-dependent cytotoxic and cell-cycle effects of* T. peruviana *cardenolides.* MCF-7 breast cancer cells were treated with convallatoxin (CT) or peruvoside (PS), two cardiac glycosides, for 24 h (left) or 72 h (right). Cell viability (%) measured by MTT assay declines with increasing concentrations of CT or PS (note the downward-sloping dose–response curves). CT (solid line) is somewhat more potent than PS (dashed line), especially at 72 h (IC_{50} ≈5 nM for CT vs ~9 nM for PS). Longer exposure (72 h) shifts the dose–response to the left, reflecting greater cytotoxicity over time. In parallel, CT and PS significantly inhibited MCF-7 colony formation and induced G_0/G_1 cell cycle arrest (not shown in the graph), corroborating their anti-proliferative mechanism. Data are mean ± SE, $p<0.05$. *(Adapted from Kaushik et al., 2017.)*

5. Oxidative Stress and Other Mechanisms:

Another feature of the anticancer activity of T. peruviana is its interaction with the oxidative stress. Interestingly, it is found that despite many anticancer agents killing by causing oxidative stress on tumor cells, T. peruviana extracts themselves are antioxidant, and could enhance antioxidant defenses of cells - either protecting normal cells or causing cell death via redox modulation. The same effect was observed in a murine study conducted in vivo (discussed below), where T. peruviana fruit extract treatment was shown to significantly decreased tumor-associated oxidative damage, even when assessed as decreased levels of malondialdehyde (lipid peroxidation) and recovery of antioxidant enzymes (superoxide dismutase, catalase, glutathione). This indicates that the extract may enhance intrinsic antioxidant mechanisms hence averting oxidative stress on normal tissues but still inducing the destruction of cancer cells. Conversely, some T. peruvinae are not found to function mostly through excessive production of reactive oxygen species (ROS) in cancer cells. Feng et al. observed that the treatment with 24 h peruvoside did not alter the ROS levels in leukemia cells significantly, neither did it interfere with mitochondrial membrane potential, which is a characteristic of non-acute oxidative bursts of apoptosis. Instead, the lethality of peruvoside was associated less with unselective ROS damage and more with specific signaling perturbations (e.g. pro-survival pathway inhibition). However, the flavonoids and phenolics found in T. peruviana (including chlorogenic acid and rutin) may also be responsible of cytotoxicity by pro-oxidant action in cancer cells or priming cells to oxidative apoptotic signals. It is not clear how exactly the redox-related pathway is triggered, but T. peruviana certainly modulates oxidative stress responses as a component of its anticancer toolkit.

In addition to apoptosis, cell cycle, and redox effects, T. peruviana agents have an effect on a number of other cancer-related pathways. The following table (Table 2) indicates the areas of important molecular mechanisms. It is important to note that peruvoside and other cardenolides are known to be Na + /K +-ATPase inhibitors and are able to cause downstream signaling events in cancer cells. NF-kB-mediated transcription, calcium signaling, and pro-death pathways have been found to be inhibited in tumor cells by an inhibition of Na+/K+ pumps by cardiac glycosids (and leading to cardiac tissue toxicity). Recent high-throughput research has developed these pathway effects. The study by Reddy et al. (2020) revealed that the Wnt/b-catenin pathway was downregulated due to the treatment with peruvoside, manifested in the reduced levels of b-catenin and the transcriptional targets (cyclin D1 and c-Myc). This is noteworthy, with Wnt/b-catenin promoting proliferation and stemness in most cancers. Moreover, peruvoside suppressed the pro-survival PI3K/AKT/mTOR pathway - an activity which not only prevents death by autophagy but also prevents autophagy (as cancer cells occasionally employ autophagy to avoid death). In fact, autophagic flux was depleted as markers of autophagy such as p62 were not significantly altered with treatment. The extract and compounds also regulate growth factor signaling, as an example, Lai et al. (2022) demonstrated that peruvoside could serve as a new Src kinase inhibitor, inhibiting Src and EGFR signaling in non-small cell lung cancer and consequently, decreasing cell migration and invasion. Convallatoxin and peruvoside inhibited epithelial-mesenchymal transition (EMT) markers (e.g. phosphorylated EGFR, vimentin, Slug) and Akt phosphorylation in breast cancer cells, which are associated with a low metastatic potential. All these varied molecular activities - encompassing regulatory elements of the apoptotic process, cell cycle proteins, and kinases as well as transcription factors - highlight that the anticancer activity of T. peruviana is multifactorial. The constituents of the plant seem to strike more than one magic bullet in that they may strike several targets on the cellular levels, which can be beneficial in defeating the redundancy and resistance mechanisms of cancer.

Table 2: Mechanisms of Anticancer Action of *T. peruviana*

Mechanistic Aspect	Evidence in *T. peruviana* Studies	Representative References
Apoptosis Induction (Intrinsic) – mitochondrial pathway	*T. peruviana* fruit extract causes DNA fragmentation (laddering) and apoptotic nuclear morphology in cancer cells. Seed glycoside activates caspase-9 and - 3, increases Bax/Bcl-2 ratio, triggering intrinsic apoptosis. PARP cleavage observed in treated cells.	Ramos-Silva et al. 2017; Cheng et al. 2016; Feng et al. 2016.
Apoptosis Induction (Extrinsic) – death receptor pathway	Flower extract enhances TNF-α/TRAIL-induced death via extrinsic pathway: amplifies caspase-8 activation and BID cleavage. Peruvoside directly activates caspase-8 in leukemia cells. Downregulation of anti- apoptotic proteins (Bcl-x_L, XIAP, survivin, Mcl-1) lowers resistance to death-receptor signals.	Managit et al., 2017; Feng et al., 2016.
Cell Cycle Arrest (Anti-proliferative effect)	*T. peruviana* compounds halt cell cycle progression: G_0/G_1 arrest in MCF-7 breast cancer (↓Cyclin D1, CDK4); G_2/M arrest in gastric and leukemia cells (↑p21^Cip1^). Arrest leads to reduced proliferation and primes cells for apoptosis.	Kaushik et al., 2017; Cheng et al., 2016; Feng et al., 2016.
Oxidative Stress Modulation	In tumor-bearing mice, *T. peruviana* extract decreases lipid peroxidation and restores SOD, catalase, GSH levels, indicating enhanced antioxidant defenses. Peruvoside does not acutely raise ROS in cancer cells (no significant ROS or mitochondrial membrane potential change). Flavonoid content (rutin, etc.) may scavenge radicals or induce mild oxidative stress to trigger apoptosis. Net effect can protect normal cells from oxidative damage while promoting tumor cell death.	Haldar et al., 2015; Feng et al., 2016; Al-Rajhi et al., 2022.
Anti- migration/Invasion (Anti-metastatic)	Fruit extract significantly inhibits cancer cell motility in wound-healing assays (delayed or no wound closure vs control). Peruvoside and convallatoxin suppress	Ramos-Silva et al. 2017; Kaushik et
Mechanistic Aspect	Evidence in *T. peruviana* Studies	Representative References

	migration and invasion of breast cancer cells in Transwell assays. These effects accompany downregulation of EMT markers (β-catenin, vimentin) and Src-EGFR signaling, reducing metastatic potential.	al., 2017; Lai et al. 2022.
Inhibition of Survival Pathways (Signal transduction)	*T. peruviana* cardenolides modulate multiple signaling networks: **Wnt/β-catenin** pathway inhibition (↓β-catenin, Cyclin D1, c-Myc) leads to reduced proliferation. **PI3K/Akt/mTOR** pathway inhibition prevents survival signaling and autophagy (extract-treated cells show decreased p-Akt and unchanged p62, indicating autophagy blockage). **MAPK/ERK** changes: peruvoside downregulates MEK1 and differentially affects ERK (context-dependent). **NF-κB and JAK-STAT** pathways are downregulated by peruvoside, lowering expression of downstream proliferation and anti-apoptotic genes. Additionally, peruvoside directly inhibits Src kinase activity, thus attenuating Src-driven pathways (e.g. FAK, EGFR) that promote growth and invasion.	Reddy et al., 2020; Kaushik et al., 2017; Lai et al., 2022.

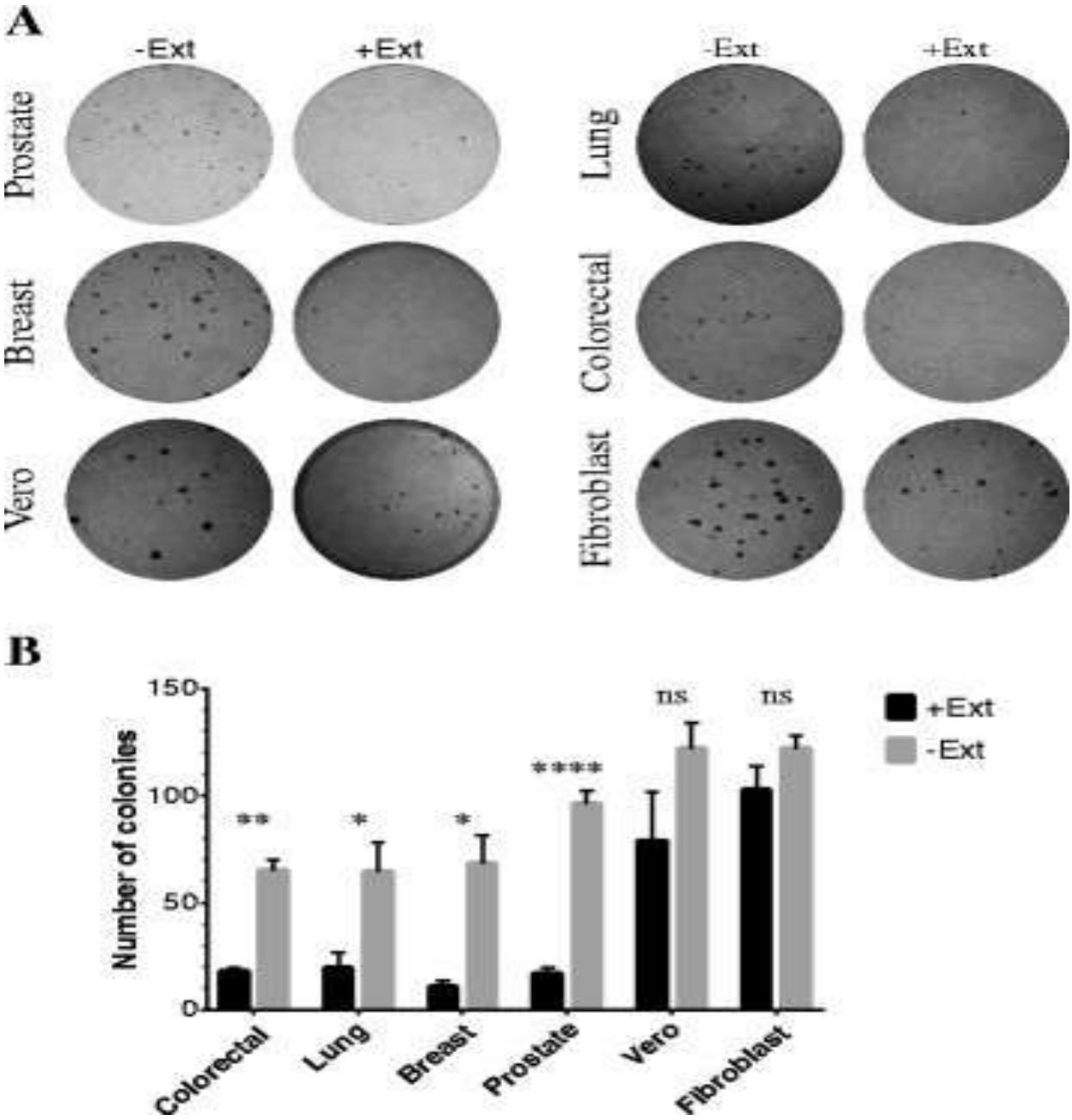


Figure 2: *In vivo antitumor efficacy of* T. peruviana *extract in mice.* Swiss albino mice bearing Ehrlich ascites carcinoma (EAC) were treated with methanolic *T. peruviana* fruit extract (METP) at 50 mg/kg or 100 mg/kg, compared to untreated controls. *(A)* Tumor volumes were markedly lower in extract-treated groups than in controls (e.g. ~2.9 mL at 50 mg/kg and ~1.3 mL at 100 mg/kg vs. ~3.6 mL in controls). Viable tumor cell counts in ascitic fluid dropped, while non-viable (dead) cell percentages rose dramatically with treatment (to ~18.6% and 68.1% at 50 and 100 mg/kg, respectively, vs. ~4.4% in controls). *(B)* Kaplan–Meier survival analysis indicated prolonged survival in *T. peruviana*-treated mice (increase in lifespan by ~49% at 50 mg/kg and ~84% at 100 mg/kg). Treated mice also showed restored hematological indices and near-normal serum chemistry, suggesting reduced tumor burden and systemic toxicity. The extract significantly decreased lipid peroxidation levels in tumor-bearing mice and boosted antioxidant enzymes (catalase, SOD, GSH), implicating enhancement of the host's oxidative defense. *(Data summarized from Haldar et al., 2015.)*

6. In Vivo Antitumor Studies:

Despite being outnumbered by in vitro studies, in vivo research data available support the anticancer potential of T. peruviana. An interesting case study was that by Haldar et al. (2015) who tested a methanol extract of T. peruviana fruits in mice with Ehrlichs ascites carcinoma (transplantable tumor model). To them, the extract showed a significant inhibition of tumor growth and an enhancement of survival of these mice in the case of daily administration of the extract. As Figure 2 showed, mice administered with T. peruviana extract possessed significantly reduced tumor volume of ascites and tumour cell counts compared to controls, which is dose-dependent. The extract inhibited tumor volume by an average of 63 percent and median survival by an average of 84 percent compared to the control animals in the highest dose (100 mg/kg body weight). Notably, the health of treated mice improved too: hematological (red/white blood cell counts, hemoglobin) and liver enzyme concentrations that are normally disturbed by tumor load were brought nearer to normal in extract-treated groups. This indicates that not only was the progression of the tumor slowed using the extract, but it also helped to reduce the systemic toxicity caused by tumors. The in vivo study, mechanistically, supported the effect of oxidative stress modulation - treated mice exhibited much reduced oxidative damage (decreased lipid peroxides) and increased antioxidant enzyme activities in the blood and tissues. The authors was able to attribute the antitumor effect to the likely augmentation of endogenous antioxidant responses, in addition to the direct cytotoxic effect of tumor cells (IC50 of the extract in EAC cells was about 58.5 ug/mL in vitro). It is important to note that these doses were tolerated well by the mice without any fatal toxicity at the effective dosage of antitumor therapy - this is very important since T. peruvina has been found to be cardiotoxic. This in vivo proof-of-concept study gives hopeful results that T. peruviana extracts can have anti-cancer effects in entire organisms, lessening tumor burden and enhancing survival without causing overt host toxicity in the situation of an adequate dose.

Outside this mouse model, historic and anecdotal evidence suggests that T. peruvina has anticancer effects in vivo (e.g. it was used in folk medicine in the treatment of cancers or tumours). Nevertheless, no controlled clinical research in humans has been conducted so far. The toxicity of the plant, which is mainly attributed to its cardiac glycosides, is a problem of clinical translation - therapeutic dosing should be done with caution to prevent cardiotoxic side effects (arrhythmias, etc.). Chemical modifications of cardiac glycosides have also been attempted or nanocarriers have been used to alter the safety profile of cardiac glycosides as anticancer agents. As an example, El-Sawi et al. (2020) developed T. peruviana leaf extracts into nanoemulsions to improve delivery: in vitro cytotoxic activity of an ethanolic extract of the plant against HepG2 liver cancer improved passive to active (e.g. the IC50 of the ethanolic extract decreasing to an active level of around 3.6 ug/mL after nano-formulation). These measures could contribute to an increase in the therapeutic window. In medicine, cardiac glycosides such as digoxin (of foxglove) and oleandrin (of Nerium oleander, a relative of T. peruviana) have been explored as having anticancer effects, and some epidemiological effects and early trials have indicated some weak anti-tumor effects. Peruvoside is still not under clinical trial, but it has strong preclinical activity (and some selectivity against cancer cells) and is an attractive candidate drug to be developed further, perhaps as an adjunct or in combination.

7. Conclusions

Thevetia peruviana is a plant of high interest in the field of oncology, and a variety of bioactive compounds with strong anticancer effects are obtained. There is extensive in vitro evidence to support that extracts and isolated constituents (particularly cardiac glycosides) of T. peruviana would be effective in killing cancer cells through induction of apoptosis (by both intrinsic mitochondrial and extrinsic death-receptor) pathways), arresting cell cycle progression, inhibiting migration/invasion of cancer cells and regulating cell survival and stress-response pathways. These effects have been proved in a wide range of cancer types - such as leukemias, and carcinoma of the breast, prostate, colon, lung, cervix and liver, frequently in very low concentrations. The anticancer

activity is further supported by in vivo experiments in tumor bearing mice where tumor regression and prolonged survival with extract treatment are observed with systemic restoration of antioxidant balance and low levels of toxicity at dose-appropriate levels. T. peruviana has a complex arsenal of anticancer components, which in a mechanistic manner act on a wide range of molecular targets: caspases and Bcl-2 family proteins, signaling kinases (SRC, Akt, ERK), and transcriptional networks (NF-kB, b-catenin) leading to the ultimate effect of cancer cell death and growth inhibition. This multi-targeted process is beneficial in terms of combating the complexity of cancer, but requires cautious research in order to make it safe since the plant is poisonous.

In future perspectives, T. peruviana can be viewed as a paradox of poisons and curing- its cardiotoxic glycosides might be isolated and used as useful chemotherapeutic agents in case of maximizing their selective and delivery. The further study ought to be conducted on bio-guided fractionation to determine the most active anticancer compounds in T. peruviana, structure-activity relationship to minimize the cardiotoxicity, and advanced drug delivery systems (nanoparticles, prodrugs) to deliver the drugs to the tumor but not to the heart. Further in vivo efficacy studies and eventually clinical studies are also justified to translate such promising results. Overall, it is possible to conclude that the cumulating evidence during the past decade presents Thevetia peruviana as a promising source of anticancer molecules, with a detailed view on the mechanistic understanding of the induction of apoptosis, regulation of oxidative stress, cell cycle arrest, and inhibition of signaling. In the framework of the strict development, this decorative lucky nut plant could bring new treatment methods - transform a dangerous poison into a weapon against cancer.

References (APA 7th ed.):

Al-Rajhi, A. M. H., Yahya, R., Abdelghany, T. M., Fareid, M. A., Mohamed, A. M., Amin, B. H., & Masrahi, A. S. (2022). *Anticancer, anticoagulant, antioxidant and antimicrobial activities of Thevetia peruviana latex with molecular docking of antimicrobial and anticancer activities*. Molecules, 27(10), 3165. https://doi.org/10.3390/molecules27103165

Cheng, H.-Y., Tian, D.-M., Tang, J.-S., Shen, W.-Z., & Yao, X.-S. (2016). *Cardiac glycosides from the seeds of Thevetia peruviana and their pro-apoptotic activity toward cancer cells*. Journal of Asian Natural Products Research, 18(9), 837–847. https://doi.org/10.1080/10286020.2016.1170814

El Gizawy, H. A., El Zanaty, S. A., El Ghaly, E. M., & Seif-Eldein, N. A. (2023). *Thevetia peruviana leaves, HPLC profile, isolation, and in vitro cytotoxicity*. RSC Advances, 13(18), 12072–12079. https://doi.org/10.1039/D3RA00588G

El-Sawi, S. A., Maamoun, A. A., Salama, A. H., & Farghaly, A. A. (2020). *Chemical profiling of Thevetia peruviana leaves cytotoxic active extracts enhanced by microemulsion formulation*. Bulletin of the National Research Centre, 44(93), 1–12. https://doi.org/10.1186/s42269-020-00339-3

Feng, Q., Leong, W. S., Liu, L., & Chan, W. I. (2016). *Peruvoside, a cardiac glycoside, induces primitive myeloid leukemia cell death*. Molecules, 21(4), 534. https://doi.org/10.3390/molecules21040534

Haldar, S., Karmakar, I., Chakraborty, M., Ahmad, D., & Haldar, P. K. (2015). *Antitumor potential of Thevetia peruviana on Ehrlich's ascites carcinoma-bearing mice*. Journal of Environmental Pathology,

Toxicology and Oncology,34(2), 105–113. https://doi.org/10.1615/JEnvironPatholToxicolOncol.2015012604

Kaushik, V., Azad, N., Yakisich, J. S., & Iyer, A. K. V. (2017). *Antitumor effects of naturally occurring cardiac glycosides convallatoxin and peruvoside on human ER+ and triple-negative breast cancers*. Cell Death Discovery, 3, 17009. https://doi.org/10.1038/cddiscovery.2017.9

Lai, Y., Chang, H., Chen, H., Chang, G., & Chen, J. J. W. (2022). *Peruvoside is a novel Src inhibitor that suppresses NSCLC cell growth and motility by downregulating multiple Src-EGFR-related pathways*. American Journal of Cancer Research, 12(6), 2576–2593. PMID: 35812056.

Managit, C., Sakurai, H., & Saiki, I. (2017). *Ethanolic extract of Thevetia peruviana flowers enhances TNF- α and TRAIL-induced apoptosis of human cervical cancer cells via intrinsic and extrinsic pathways*. Oncology Letters, 13(4), 2791–2798. https://doi.org/10.3892/ol.2017.5748

Ramos-Silva, A., Tavares-Carreón, F., Figueroa, M., De la Torre-Zavala, S., Gastelum-Arellanez, A., Rodríguez-García, A., … & Campos, A. (2017). *Anticancer potential of Thevetia peruviana fruit methanolic extract*. BMC Complementary and Alternative Medicine, 17(1), 241. https://doi.org/10.1186/s12906-017-1751-6

Reddy, D., Kumavath, R., Tan, T. Z., Ampasala, D. R., & Kumar, A. P. (2020). *Peruvoside targets apoptosis and autophagy through MAPK, Wnt/β-catenin and PI3K/AKT/mTOR signaling pathways in human cancers*. Life Sciences, 241, 117147. https://doi.org/10.1016/j.lfs.2019.117147

Silalahi, M. (2022). *Thevetia peruviana (Pers.) K. Schum.: Poisonous and its potential*. Magna Scientia Advanced Research and Reviews, 6(1), 31–37. https://doi.org/10.30574/msarr.2022.6.1.0064

Tian, D.-M., Cheng, H.-Y., Jiang, M.-M., Shen, W.-Z., Tang, J.-S., & Yao, X.-S. (2016). *Cardiac glycosides from the seeds of Thevetia peruviana*. Journal of Natural Products, 79(1), 38–50. https://doi.org/10.1021/acs.jnatprod.5b00574