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ANTICANCER POTENTIAL OF *BARLERIA STRIGOSA WILLD.* (ACANTHACEAE): A COMPREHENSIVE REVIEW OF PHYTOCHEMISTRY, IN VITRO EVIDENCE, AND MECHANISTIC INSIGHTS

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ABSTRACT

Background: *Barleria strigosa Willd.* is a relatively under-explored medicinal plant of the family Acanthaceae, distributed across tropical Asia and used in traditional practice for fever, inflammation, and detoxification. The genus *Barleria* is a recognised reservoir of iridoid glycosides, phenylethanoid glycosides, flavonoids and terpenoids—chemical classes repeatedly linked to antiproliferative activity—yet the anticancer profile of *B. strigosa* specifically has only recently begun to be characterised.

Objective: To consolidate and critically appraise the available experimental evidence on the anticancer/antiproliferative activity of *B. strigosa*, to map the phytoconstituents plausibly responsible, and to identify the mechanistic and translational gaps that should direct future work.

Summary of evidence: Three independent in vitro investigations provide direct signals of activity. Leaf extracts show concentration-dependent cytotoxicity and anti-migration against oral squamous carcinoma (CLS-354/WT) cells with morphological hallmarks of apoptosis; butanolic and methanolic leaf extracts are cytotoxic to HeLa (cervical), MCF-7 (breast) and P-388 (murine leukaemia) lines; and a 2025 study demonstrated that ethanolic *B. strigosa* extract strongly reduced prostate cancer (PC-

3) viability, induced DNA damage and G1 arrest, and activated peripheral blood mononuclear cells against PC-3 through cytokine gene upregulation. Activity is attributed chiefly to verbascoside, barlerin/acetylbarlerin, related iridoids, and flavonoids. Significantly, the same 2025 investigation highlighted a selectivity warning by identifying detectable toxicity of B. strigosa toward certain normal cells (hepatocytes and prostate epithelium).

Conclusion: *Barleria strigosa* is a promising but early-stage anticancer candidate. Current data are wholly in vitro, mechanistically incomplete, and complicated by normal-cell toxicity signals. Bioassay-guided isolation validated molecular-pathway studies, selectivity indexing, and in vivo confirmation are the essential next steps before any therapeutic claim can be entertained.

KEYWORDS: Barleria strigosa; Acanthaceae; anticancer; antiproliferative; verbascoside; barlerin; iridoid glycosides; apoptosis; ethnopharmacology

1. INTRODUCTION

Cancer remains one of the leading causes of death worldwide, and the limitations of conventional chemotherapy—systemic toxicity, poor selectivity between malignant and normal cells, and the emergence of resistance—continue to drive the search for safer, selectively acting agents. Plant-derived natural products have historically been, and remain, a productive source of such leads; a substantial proportion of clinically used anticancer drugs are either natural products or derived from them. Within this landscape, plants used in traditional medicine offer a rational starting point because their long human use provides preliminary signals of tolerability and biological activity.

The genus *Barleria* L. (Acanthaceae) is the third-largest genus in its family and comprises roughly 300 species distributed across tropical and subtropical regions. Several species—*B. prionitis*, *B. cristata*, *B. lupulina*, *B. grandiflora* and *B. longiflora*—have been studied for antioxidant, anti-inflammatory, antimicrobial, antidiabetic and, increasingly, anticancer properties. These activities are consistently ascribed to a characteristic secondary-metabolite palette: iridoid and phenylethanoid glycosides (notably barlerin, acetylbarlerin and verbascoside), flavonoids, phenolic acids, quinones and terpenoids.

Barleria strigosa Willd. is comparatively neglected within this well-studied genus. Native to tropical Asia and used traditionally for fever, influenza, nosebleed and as an antidote in detoxification, it shares the genus' phytochemical signature—phenylethanoid glycosides, iridoid glycosides, phenolic acids and flavonoids have all been isolated from it—but until recently attracted very little pharmacological attention, and almost none directed at cancer. Over the last few years, however, three lines of in vitro evidence have converged to suggest genuine antiproliferative potential, making a focused synthesis both timely and necessary.

This review therefore aims to (i) summarise the botanical and ethnomedicinal background of *B. strigosa*; (ii) catalogue its anticancer-relevant phytochemistry; (iii) critically appraise the direct

experimental evidence for its antiproliferative activity; (iv) reconstruct the plausible molecular mechanisms from both extract-level and isolated-compound data; (v) place *B. strigosa* in comparative context within the genus; and (vi) delineate the safety concerns and research gaps that must be addressed before translational claims are justified.

2. LITERATURE REVIEW

This is a *narrative* review. Peer-reviewed literature was retrieved from PubMed, Scopus, ScienceDirect, SpringerLink and Google Scholar using combinations of the terms Barleria strigosa, anticancer, antiproliferative, cytotoxic, apoptosis, verbascoside, barlerin and iridoid glycosid. Because direct reports on *B. strigosa* are few, the evidence base was deliberately widened in two controlled ways: first, to isolated-compound studies on the specific constituents identified in *B. strigosa*; and second, to congeneric *Barleria* species, used only as supporting/comparative context and clearly labelled as such. Readers should note that a formal PRISMA-based systematic review was not performed; findings are qualitative and interpretive rather than quantitatively pooled.

3. BOTANICAL PROFILE ETHNOMEDICINAL USE

Barleria strigosa Willd. is a perennial herb/undershrub of the Acanthaceae, occurring in tropical Asia including Thailand, Laos, Myanmar and parts of India. In north Indochina it is one of four commonly cited *Barleria* species alongside *B. prionitis*, *B. lupulina* and *B. cristata*. Traditionally, *B. strigosa* has been employed for the management of fever and influenza, to arrest nosebleed, and as an antidote in the detoxification of poisons. Notably, in parts of Thailand, boiled *Barleria* leaves—including those of *B. strigosa*—have featured in anecdotal folk use by local herbalists as a supportive measure for cancer patients. Such anecdotal reports are not evidence of efficacy, but they do provide an ethnopharmacological rationale that has motivated recent laboratory screening.

4. ANTICANCER-RELEVANT PHYTOCHEMISTRY OF B. STRIGOSE

Historically, phytochemical study of *B. strigosa* was limited to a small number of reports describing phenylethanoid glycosides, iridoid glycosides, phenolic acids and flavonoids. Early work by Kanchanapoom and colleagues characterised phenylethanoid and iridoid glycosides from the Thai plant, and later Prapalert and colleagues isolated two phenylethanoid glycosides, parvifloroside A and B. Deepak and colleagues subsequently profiled medicinally active flavonoids, phenolic compounds and terpenoids and reported associated antioxidant activity.

The most detailed profile to date comes from a 2023 UPLC–ESI–QTOF–MS study, which tentatively identified 82 compounds in a hydrophilic leaf extract. Terpenoids dominated (38 detected, including 15 diterpenes such as (+)-totarol, longikaurin A and hautriwaic acid, plus diterpene lactones/glycosides). The extract was also rich in iridoid glycosides (e.g., dihydrocatalpol, verproside, harpagoside, catalposide, durantoside I), flavonoids (including a hyperin derivative, plumerubroside, sophoranochromene and an epicatechin dimer), phenolic glycosides, a coumarin (daphnoretin methyl

ether), a naphthoquinone and an isoferulic-acid phenylpropanoid. A 2025 GC–MS/HPLC study independently confirmed two constituents of particular anticancer interest—**verbascoside** and **barlerin**—in *B. strigosa* leaf, with stigmasterol reported as its dominant relative constituent by that method. Several of these classes—phenylethanoid glycosides, iridoids, flavonoids and specific terpenoids—are precisely the constituents with independent anticancer literature, and thus form the mechanistic bridge discussed in Section 7.

Table 01. Selected anticancer-relevant constituents reported from B. strigose.

Compound / class	Chemical class	Anticancer relevance (evidence source)	Ref.
Verbascoside (acteoside)	Phenylethanoid glycoside	Confirmed in B. strigosa leaf; independently pro-apoptotic in breast (4T1) and other tumour cells via Bax/Bcl-2 and caspase-3	6, 8, 16
Barlerin / acetylbarlerin	Iridoid glycoside	Confirmed in B. strigosa leaf; reported anticancer & chemopreventive (NQO1 induction) activity in the genus	6, 8, 17
Iridoid glycosides (verproside, catalposide, harpagoside, durantoside I, dihydrocatalpol)	Iridoid glycosides	Class detected by UPLC-QTOF-MS; congeneric iridoids (prionitosides) cytotoxic to A549	6, 14
Flavonoids (hyperin deriv., plumerubroside, epicatechin dimer)	Flavonoids	Detected in leaf; flavonoids are a broadly established antiproliferative class	6, 7
Diterpenes/terpenoids (totarol, longikaurin A, hautriwaic acid)	Terpenoids	Major class in leaf; several kaurane/abietane diterpenes are cytotoxic in other taxa	6
Stigmasterol	Phytosterol	Reported as dominant GC-MS constituent; phytosterols show antiproliferative signals in congeners (e.g., B. longiflora docking)	8, 15

Reference numbers correspond to the reference list. "Congeneric" evidence is supportive/comparative, not direct evidence for B. strigosa.

5. Direct experimental evidence for anticancer activity

Three primary in vitro studies constitute the current direct evidence base. They are summarised in Table 2 and detailed below.

5.1 Oral squamous Carcinoma (CLS- 354/WT)

In the 2023 study by Lei and colleagues, hydrophilic (BSTR) and lipophilic (LSB) ethanolic leaf extracts were tested against the oral squamous carcinoma line CLS-354/WT by MTT assay. Both extracts produced concentration-dependent cytotoxicity, with BSTR reaching approximately 90% cytotoxicity between 400 and 1600 µg/mL and a CC^{50} of about 234 µg/mL, modestly more potent than LSB ($CC^{50} \approx 313$ µg/mL). Beyond simple viability loss, both extracts significantly inhibited

cancer-cell migration in a scratch assay, and DAPI nuclear staining revealed condensed, fragmented chromatin—morphological hallmarks indicating that cell death proceeded predominantly through apoptosis rather than necrosis. The same extracts were comparatively biocompatible with non-cancer macrophage (RAW 264.7) cells (IC^{50} > 1000 µg/mL), and also displayed antioxidant, α-glucosidase/α-amylase-inhibitory, anti-inflammatory (nitric-oxide-lowering) and antibacterial activities, situating the anticancer signal within a broad pharmacological profile.

5.2 Cervical (heLa), Breast (MCF-70) and murine Leukaemia (P-388)

The earliest direct cytotoxicity data derive from Manapradit and colleagues (2015). Among solvent leaf extracts of *B. strigosa*, the butanolic fraction exhibited the highest cytotoxicity against HeLa (cervical) and MCF-7 (breast) carcinoma cells, while the methanolic leaf extract was cytotoxic to the P-388 murine leukaemia line with a 50% cytotoxic concentration (CC^{50}) of approximately 414 µg/mL. These findings—though based on crude extracts and relatively high concentrations—established that *B. strigosa* cytotoxicity is not restricted to a single tumour type and is solvent-fraction dependent, implicating semi-polar constituents (glycosides, flavonoids, phenolics) in the effect.

5.3 Prostate cancer (PC-3): cytotoxicity, DNA damage and immunomodulation

The most mechanistically informative study is that of Kaewdaungdee and colleagues (2025), which compared six *Barleria* leaf extracts. *B. strigosa* extract exerted strong cytotoxicity against prostate cancer PC-3 cells, reducing viability to below 20%, and was classed as selectively toxic toward the cancer line. Using comet assay and flow cytometry, the extract was shown to induce significant DNA damage (elevated olive tail moment) in PC-3 while sparing that damage in immune (PBMC) cells, and to arrest the PC-3 cell cycle in G1 phase, suppressing progression through S and G2–M.

A distinctive and translationally interesting finding was **immunomodulation**. When peripheral blood mononuclear cells (PBMCs) were pre-activated for 48 h with *B. strigosa* extract and then co-cultured with PC-3 cells, cancer viability fell to roughly 73%, indicating an indirect, immune-mediated route of tumour suppression in addition to direct cytotoxicity. Consistently, *B. strigosa* upregulated the cytokine genes IL-2, IL-10 and IL-12 in PBMCs (relative expression several-fold above control), although—unlike the more potent *B. siamensis*—it did not raise IFN-γ. The authors attributed the shared genus-level activity chiefly to the common metabolites verbascoside and barlerin, and noted that some *Barleria* extracts (including *B. strigosa*) showed greater cancer-selectivity than cisplatin, which was toxic to normal cells as well.

Table 2. Summary of direct in vitro anticancer studies on *B. strigose.*

Study (year)	Extract / part	Cancer model	Key findings	Ref.
Manapradit et al. (2015)	Butanolic & methanolic leaf	HeLa, MCF-7, P-388	Butanolic extract most cytotoxic to HeLa & MCF-7; methanolic extract CC50 ≈ 414 µg/mL vs P-388 leukaemia	3
Lei/Olatunji et al. (2023)	Hydrophilic (BSTR) & lipophilic (LSB) ethanolic leaf	Oral squamous carcinoma CLS-354/WT	Dose-dependent cytotoxicity (BSTR CC50 ≈ 234 µg/mL); anti-migration; apoptotic nuclei (DAPI); biocompatible with RAW 264.7	6
Kaewdaungdee et al. (2025)	Ethanolic leaf	Prostate PC-3 (+ normal PBMC/HPrEC/THLE-3)	PC-3 viability <20%; DNA damage; G1 arrest; PBMC activation & cytokine (IL-2/-10/-12) upregulation; selective vs cisplatin but some normal-cell toxicity	2

CC50/IC50 = concentration reducing viability by 50%. Values are for crude extracts and are not directly comparable across studies due to differing solvents, cell lines and assay conditions.

6. Plausible and molecular mechanisms

Although no single study has fully dissected the anticancer mechanism of *B. strigosa*, the extract-level observations and the known pharmacology of its constituents converge on several complementary pathways.

- **Apoptosis induction.** Chromatin condensation and nuclear fragmentation (DAPI) in CLS-354/WT cells, and annexin-V/PI-defined apoptosis with DNA fragmentation (comet) in PC-3, indicate programmed cell death as a primary mode. This is congruent with verbascoside's documented ability to raise the Bax/Bcl-2 ratio and activate caspase-3 in tumour cells.
- **Cell-cycle arrest.** B. strigosa extract arrested PC-3 in G1 phase, curtailing S-phase DNA synthesis and G2–M division—a checkpoint-level brake on proliferation.
- **Genotoxic stress selective to tumour cells.** Elevated olive tail moment in PC-3 but not in PBMCs suggests DNA-damage that is preferentially inflicted on, or less well repaired by, malignant cells.
- **Anti-migration/anti-metastatic signal**. CLS-354/WT significantly inhibited scratch-wound closure, suggesting interference with cancer cell motility, an essential determinant for invasion and metastasis .
- **immunomodulation.** However, the PBMC activation and overexpression of anticancer related cytokine genes (IL-2, IL-10, and IL-12) mediate cytotoxicity against PC-3 thru the immune system indirectly, unlike direct chemical death.

- **Antioxidant / anti-inflammatory context.** Strong radical-scavenging (DPPH, ABTS, FRAP) and nitric-oxide-lowering activity may contribute to chemoprevention by attenuating the oxidative-inflammatory milieu that promotes carcinogenesis.
- **Drug-metabolism (CYP450) modulation.** B. strigosa induced hepatic CYP2E1/CYP2A6 expression in THLE-3 cells; because CYP enzymes govern xenobiotic activation and clearance, such modulation could influence both procarcinogen handling and herb–drug interactions—an effect requiring careful evaluation.

7. Constituent- Level Evidence Bridging to *B. srigosa*

7.1 Verbascoside (acteoside)

Verbascoside, a caffeoyl-phenylethanoid glycoside confirmed in *B. strigosa* leaf, is among the best-supported anticancer constituents of the genus. In breast cancer 4T1 cells it inhibited proliferation ($IC^{50} \approx 117$ μM) while sparing normal HEK293T cells, and mechanistically increased the pro-apoptotic Bax/Bcl-2 ratio, activated caspase-3, and modulated the TLR4/MyD88/NF-κB axis. Its presence therefore offers a direct molecular rationale for the apoptotic phenotype observed with *B. strigosa* extracts.

7.2 Barlerin and acetylbarlerin

Barlerin (shanzhiside-type iridoid glycoside) and its acetyl derivative, both reported across *Barleria* species and confirmed in *B. strigosa*, carry documented anticancer and chemopreventive activity. Acetylbarlerin from *B. trispinosa*, for example, was shown to induce the chemoprotective phase-II enzyme NQO1 while attenuating LPS-driven inflammation, consistent with a carcinogenesis-blocking role rather than only direct cytotoxicity.

7.3 Iridoid glycosides

Iridoid glycosides are a defining chemical feature of the genus. Congeneric evidence is instructive: prionitosides and related iridoids isolated from *B. prionitis* exhibited cytotoxicity against A549 lung carcinoma cells alongside nitric-oxide inhibition. The rich iridoid inventory of *B. strigosa* (verproside, catalposide, harpagoside, durantoside I, dihydrocatalpol) is thus a plausible contributor to its antiproliferative signal, warranting bioassay-guided isolation.

7.4 Flavonoids and terpenoids

Flavonoids are a broadly validated anticancer class, and their presence in *B. strigosa* leaf aligns with the cytotoxicity attributed to semi-polar fractions. The terpenoid-dominated profile—particularly diterpenes—adds further candidate scaffolds; in the congener *B. longiflora*, phytosterols such as stigmasterol emerged as favourable in-silico binders in an anticancer screen against A549, mirroring stigmasterol's prominence in *B. strigosa*.

8. Comparative Context within the Genus Barleria

Placing *B. strigosa* against its relatives clarifies both its promise and its liabilities. In the six-species 2025 comparison, *B. cristata*, *B. siamensis* and *B. strigosa* were the strongest anticancer candidates, whereas *B. prionitis* and *B. repens* were largely inactive (correlating with their low verbascoside/barlerin content), and *B. lupulina* was potent but non-selective (toxic to normal cells). Among the active trio, *B. siamensis* was the most favourable—high potency, clean normal-cell profile, and strong immunomodulation—while *B. strigosa*, despite strong PC-3 killing, showed the weakest cancer-selectivity of the three because of measurable normal-cell toxicity (see Section 9). Independent congeneric anticancer reports—*B. cristata* phenolics as quinone-reductase inducers, *B. grandiflora* leaf extract against A-549 and Dalton's lymphoma, and *B. prionitis* iridoids against A549—reinforce that anticancer activity is a genus-wide but chemotype-dependent trait.

9. Safety and Selectivity Considerations

A credible appraisal must foreground the toxicity signal. In the 2025 study, *B. strigosa* extract was not cleanly selective: it reduced normal prostate epithelial (HPrEC) viability to about 46% at 100 μg/mL and inhibited non-tumoral hepatocyte (THLE-3) growth (viability ≈ 58%), inducing DNA damage, apoptosis/necrosis and G1 arrest in the hepatocytes as well. The authors explicitly cautioned that *B. strigosa* "may not be entirely safe for use." By contrast, the 2023 CLS-354/WT study reported good biocompatibility with macrophages, and the 2025 work still classed *B. strigosa* as more cancer-selective than cisplatin. These are not contradictions so much as a reminder that selectivity is cell-line- and concentration-dependent. The induction of hepatic CYP2E1/CYP2A6 further raises the prospect of herb–drug interactions. Collectively, these findings mandate rigorous selectivity indexing and normal-cell panels in all future work, rather than reliance on cancer-cell potency alone.

10. Research Gaps and Future Directions

- **No in vivo or clinical data.** All existing evidence is in vitro; tumour-xenograft and toxicology studies are absent and are the single most important next step.
- **Crude extracts, not isolated actives.** Effects are documented for whole extracts; bioassay-guided fractionation is needed to attribute activity to specific molecules and quantify their contribution/synergy.
- **Incomplete mechanism.** Key pathways (caspase cascades, Bax/Bcl-2, p53, PI3K/AKT, NF-κB, STAT3) have been inferred but not directly measured in B. strigosa–treated cells.
- **Selectivity unresolved.** Normal-cell toxicity signals demand systematic selectivity-index determination across matched normal/tumour pairs.
- **Standardisation and quality control.** Constituent content (verbascoside, barlerin) varies with solvent, geography and season; standardised, chemically fingerprinted extracts are prerequisites for reproducibility.

- **Pharmacokinetics and herb–drug interactions.** CYP450 modulation observed in vitro must be characterised for translational safety.
- **Formulation.** Glycosidic actives such as verbascoside have bioavailability constraints that nanocarrier or prodrug strategies may need to address.

11. CONCLUSION

Barleria strigosa has moved, within a few years, from a pharmacologically obscure plant to a documented in vitro antiproliferative agent active against oral, cervical, breast, leukaemic and prostate cancer models, acting through apoptosis, cell-cycle arrest, tumour-selective DNA damage, anti-migration and immune activation. Its activity is chemically coherent, resting on well-characterised anticancer constituents—verbascoside, barlerin/acetylbarlerin, iridoids and flavonoids—shared with more-studied congeners. Yet the evidence is early-stage: exclusively cell-based, mechanistically inferential, and shadowed by genuine normal-cell toxicity and CYP-modulation signals. *B. strigosa* is best regarded not as a validated anticancer therapy but as a credible lead scaffold whose promise now hinges on bioassay-guided isolation, mechanistic confirmation, selectivity indexing and in vivo validation. Framed that way, it is a rewarding and under-exploited target for rigorous cancer-pharmacognosy research.

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