

Therapeutic Avenues of Bacteriochlorophyll Derivatives

Vikrant Arya^{1,*}, Ranjeet Kaur Parmar², Amandeep Kaur Gill³

¹Department of Pharmaceutical Sciences, Government Pharmacy College, Rakkar, Kangra, Himachal Pradesh, INDIA.

²Department of Pharmaceutical Sciences, Government Pharmacy College Kangra at Nagrota, Bagwan, Himachal Pradesh, INDIA.

³School of Pharmaceutical Sciences, CT University, Ludhiana, Punjab, INDIA.

ABSTRACT

Background: Bacteriochlorophylls are specialized bacterial pigments produced naturally in certain bacteria to convert captured light into chemical energy. These substances and their metabolic precursors (bacteriochlorophyllides) demonstrate high effectiveness in diagnosing and treating diverse medical conditions. They are particularly useful for deep-tissue cancer imaging and in tumor eradication. **Materials and Methods:** A comprehensive literature search was conducted across scientific databases, viz. Google Scholar, PubMed, SciHub, and Web of Science. **Results:** BChl a conjugates like Redaporfin®, Tookad®, have been clinically tested in improving tumor eradication via a PDT approach. They have also been reported to be antioxidants and anti-ageing, and to exert significant activity on glioblastoma cells. BChl-ser-treated tumors result in total oxygen depletion within the tissue, highlighting their therapeutic relevance. The specific light absorption range allows for deeper penetration into necrotic tissues while maintaining low toxicity. This property facilitates effective tumour removal via photodynamic therapy by increasing photodamage in the target area. **Conclusion:** The derivatives of light-harvesting pigment, bacteriochlorophyll, exhibit diversified applications ranging from ecology to therapeutics. Notably, their conjugates were found to be pharmacologically active, particularly in the treatment of specific types of tumors. Furthermore, Bacteriochlorophyll pigments and their derivatives show significant potential in non-oncological fields.

Keywords: Bacteriochlorophyll, Bacteriochlorophyllides, Bacteriochlorophyll-c-stearyl, Chemotherapy, Drug Delivery.

Correspondence:

Dr. Vikrant Arya

Department of Pharmaceutical Sciences,
Government Pharmacy College, Rakkar,
Kangra-177043, Himachal Pradesh, INDIA.
Email: arya.vikrant30@gmail.com

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INTRODUCTION

Treating cancer remains one of the formidable challenges in the medical field for both developed and developing countries. Despite current medications and approaches, cancer seems to be expanding at an exponential rate throughout the world.^{1,2} In a 2025 report, the American Cancer Society estimated that there were 2,041,910 new invasive cancer cases in the United States during that year.³ Inherent heterogeneity and complexity of tumor biology seem to be the major obstacles in designing anti-cancer drugs.⁴ Chemotherapy resistance is another alarming concern in cancer treatment and prevention, leading to recurring symptoms.⁵ Natural products, particularly plants, offer a preventive approach in combating cancer, although discrete information, a lack of comprehensive research, and insufficient clinical data restrain us from using them as a first-line measure of cancer.⁶

Bacteria and their photosynthetic pigments have been harnessed for their anti-cancer properties.^{7,8} Bacteriochlorophyll and its associated derivatives exhibit a unique ability to target and accumulate in cancer tissues, thereby initiating anti-cancer immune responses in various types of tumors. The term bacteriochlorophylls (BChl) is derived from two words, 'bacterio' representing the bacteria, and 'chlorophyll' referring to a photosynthetic pigment. These are photosynthetic pigments present in several phototrophic bacteria, enabling organisms to perform anoxygenic photosynthesis.⁹ BChl has been explored for properties ranging from a light-harvesting pigment¹⁰ to an anti-tumor agent since its existence in 1932.¹¹ Chemically related to tetrapyrrole, BChl has structural similarity to chlorophyll, encompassing light-harvesting capabilities.¹² Generally, BChl and its associated derivatives are present in photosynthetic bacteria.¹³ However, they can also be produced synthetically through chemical reactions.¹⁴ Their presence can be seen in several phototrophs, viz., purple bacteria (*Cereibacter sphaeroides*, *Acidiphilium rubrum*), green sulfur bacteria (*Chlorobaculum tepidum*, *C. parvum*), heliobacteria (*Heliobacterium mobile*), Chloroflexus (*Chloroflexus aurantiacus*, *Roseiflexus castenholzii*), etc.¹⁵ In ecology, they play a critical role in anoxygenic photosynthesis,¹⁶ the carbon cycle,¹⁷ the sulfur metabolism in



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the case of green sulfur bacteria,¹⁸ and aid in the adaptation of phototrophic bacteria¹⁹ as well as in energy conversion.²⁰

Having the property of being photosensitizers, several BChl derivatives have been used in cancer therapy.²¹ Photochemotherapy is a treatment approach for treating cancers that involves a photosensitizer molecule/drug with light.^{22,23} Light-induced generation of reactive oxygen species by photosensitized BChl conjugates resulted in irreparable cell membrane damage, and apoptosis is the key underlying mechanism involved in tumor eradication.^{24,25} Among the seven existing BChl (BChl a to g), the associated derivatives of BChl a and c are mainly employed in cancer research. The synthetic derivatives of BChl have been harnessed in treating different types of tumors²⁶ due to their inherent ability to absorb light in the near-infrared region from 700 to over 800 nm,¹¹ which allows deeper tissue penetration and is proven effective in photodynamic therapy.²⁷ Conjugates/derivatives including BChl-T,²⁸ BChl-ser,²⁹ bacteriochlorins,³⁰ metallobacteriochlorophyll,³¹ and palladium bacteriopheophorbide³² have been employed in targeting tumour cells. Apart from therapeutic applications, BChl derivatives have wide applications in target-specific immunoassay,³³ dye-sensitized solar cells,^{34,35} heme synthesis,³⁶ and light energy harvesting simulation.³⁷ Research into these bacterial pigments involves utilizing their derivatives as diagnostic markers for *in vivo* imaging and evaluating their potential as anticancer agents. Studies often focus on their inherent ability to absorb light within the 770 to 850 nm range. Development efforts also include creating semi-synthetic or water-soluble derivatives to improve solubility and performance in vascular-targeted photodynamic therapy and other diseases. In this current review, an attempt has been made to highlight the therapeutic potential of BChl derivatives with emphasis on their potential applications in oncology.

MATERIALS AND METHODS

A comprehensive literature search was conducted across scientific databases, viz. Google Scholar, PubMed, SciHub, and Web of Science. This exploration utilised several keywords relevant to the topic, like 'bacteriochlorophyll', 'bacteriochlorophyll derivatives in tumor targeting', 'BChl in photodynamic therapy', and 'metallobacteriochlorophylls', etc. This systematic approach ensured wide coverage of the selected topic and paved the way for further investigation.

RESULTS

Early Discoveries and Structural Elucidation

Richard Willstätter, a Nobel Laureate German chemist, along with his colleagues, conducted several experiments between 1905 and 1914 and contributed to determining the empirical formula of chlorophyll.³⁸ Bacteriochlorophylls came into light when, in 1932, Cornelis Bernardus van Niel, a microbiologist, worked on purple and green sulfur bacteria, which contain this

photosynthetic pigment. He investigated the physiology and morphology of purple and green sulfur bacteria, which contain these pigments.³⁹⁻⁴¹ In 1935, Johann Hasenkamp, along with Hans Fischer, reported the isolation of bacteriochlorophyll a. Subsequently, in 1937, they proposed its chemical structure and formulation.⁴² In 1954, the enzyme-mediated conversion of bacteriochlorophyll into bacteriochlorophyllide and its determination via spectroscopy paved the way for new avenues in the BChl derivatives.⁴³ Later, in 1963, a team led by Eimhjellen, Aasmundru, and Jensen succeeded in isolating Bacteriochlorophyll b from *Rhodospseudomonas*.⁴⁴ The period between 1966 and 2012 led to more advancements in structural elucidation of the pigment family, viz. bacteriochlorophyll c, d, e, f, and g. The major chronological events in the discovery path of bacteriochlorophyll have been illustrated in Figure 1.⁴⁵⁻⁵²

Sources of Bacteriochlorophyll

Bacteriochlorophylls can be divided into two sources: natural or synthetic (Figure 2). Naturally, they are found in green sulfur bacteria, purple bacteria, and heliobacteria.⁵³⁻⁵⁶ Photosynthetic bacteria, viz. *Rhodobacter capsulatus* is reported to contain BChl a, which can be easily separated by extraction (Yurkova & Beatty, 1996).⁵⁷ Several green sulfur bacteria are known to produce BChl c, BChl d, and BChl e, which are components of their chlorosomes (light-harvesting organelles/ antenna).⁵⁸⁻⁶⁰

Photosynthetic Gram-negative purple proteobacteria, *Rhodospseudomonas* (Blastochloris), are the major source of BChl b,^{61,62} whereas green sulfur bacteria, *Chlorobium*, are a source of BChl c, d, e, f (mutant).^{63,64,51} *Heliobacteria*, a prokaryotic anoxygenic light-harvesting bacterium, contain BChl g.^{65,55} Synthetic conjugates of bacteriochlorophyll can be produced via chemical modifications to the existing structure of BChl, chemical synthesis,⁶⁶ and functional group additions.⁶⁷ Modifications of natural bacteriochlorophyll and de novo synthesis can be used to produce BChl derivatives.⁶⁸ Chemical reactions (palladium-catalyzed cross-coupling reactions, transesterification) are usually employed to transform these photosynthetic pigments into tumour-eradicating candidates.^{67,69,70}

Types of Bacteriochlorophylls

Bacteriochlorophylls (BChl) are photosynthetic pigments structurally related to plant pigment chlorophylls present in several phototrophic bacteria, which were discovered by a microbiologist, Cornelis Bernardus van Niel, in 1932.⁵² They are represented as a versatile group of chlorophyll derivatives typically found in phototrophic bacteria. Their seven key types, as represented in Figure 3, include BChl a ($C_{55}H_{74}MgN_4O_6$), BChl b ($C_{55}H_{72}MgN_4O_6$), BChl c, BChl d, BChl e, BChl f, and BChl g. These can be modelled and conjugated with various other molecules for diverse applications.^{51,52} Bacteriochlorophylls consist of chlorophyllide a, which is a biosynthetic precursor

involved in their production. BChl can be categorised mainly into two types, viz. bacteriochlorins (BChl a, b, g) and Chlorins (BChl c, d, e, f).^{48,52,71} Bacteriochlorophyllides (BChlide), characterised as esterifying alcohol-free forms of bacteriochlorophylls (BChl), are the immediate precursor to bacteriochlorophyll, are also widely used as photosensitizers in photodynamic therapy. Currently, BChlides are also identified into seven types as Bacteriochlorophyllides a to g, and their several derivatives have been used in PDT.⁵²

Therapeutic relevance

The ROS-generating capability of photosensitive molecules offers a lead in treating cancers. These photosensitizers can be activated by light to initiate ROS production, thereby targeting tumours by causing irreparable damage to cancer cells. BChl a, isolated from the purple nonsulfur bacterium (*Rhodobacter sphaeroides*), exhibits significant skin anti-ageing effects via significant improvements in procollagen production.⁷² BChl-a has also been used in eye-related conditions, particularly in corneal thinning and ectasia.⁷³⁻⁷⁵

BChl-a is successfully employed as a cytotoxic agent in non-invasive cancer treatment (sonodynamic therapy). Its capacity to generate ROS via ultrasound activation is reported to inhibit tumor growth.⁷⁶ A patent (KR101320045B1) encompassing a cosmetic formulation prepared from bacteriochlorophyll derived from purple bacteria provides moisturization and alleviates skin irritation.⁷⁷ Photobac, a photosensitizer derivative of bacteriochlorophyll-a, has been reported to exhibit cytotoxic effects in glioblastoma and associated tumors when evaluated

in several cell lines (U87, F98, C6). The drug showed desired pharmacokinetic properties, having limited toxicity when evaluated in rats and dogs, and received the status of orphan drug from the US FDA, as well as received approval for phase I human clinical trials due to its effectiveness in glioblastoma.⁷⁸ Redaporfin, a synthetic BChl a derivative, which is currently in Phase II clinical trials, demonstrated potential in treating head and neck carcinoma designed for Photodynamic therapy.^{79,80} In a study involving a BChl derivative by Pratavieira *et al.* (2021), a semi-synthetic derivative of bacteriochlorin was synthesized from *Rhodopseudomonas faecalis*, and its poor solubility was overcome by adding Trizma®. The efficacy of the resulting derivative of bacteriochlorin-Trizma (BChl-T) was evaluated in HFF-1, MCF-7 cells. The results demonstrated around 80% cell death in the breast cancer cell line (MCF-7) at concentrations above 0.5 μM .²⁸

Bacteriochlorophyll-serine, a Bacteriochlorophyll derivative synthesized via the transesterification of bacteriochlorophyll (BChl) with a non-essential amino acid (serine), has been successfully employed in PDT via modulating oxygen dynamics in melanoma-associated tumors.²⁹ The findings on the synergistic impact of hyperthermia and bacteriochlorophyll derivative (BChl-ser) were evaluated on tumor metabolism. The results highlighted disruption of tumor energy metabolism via an increase in lactate and a decrease in ATP levels, suggesting profound PDT effects in the tumor via altering microenvironmental changes.⁸¹ Nanoformulation prepared from a tetrapyrrole bacteriochlorin derivative enhances the therapeutic effects in PDT via reducing toxicity and enhancing solubility,

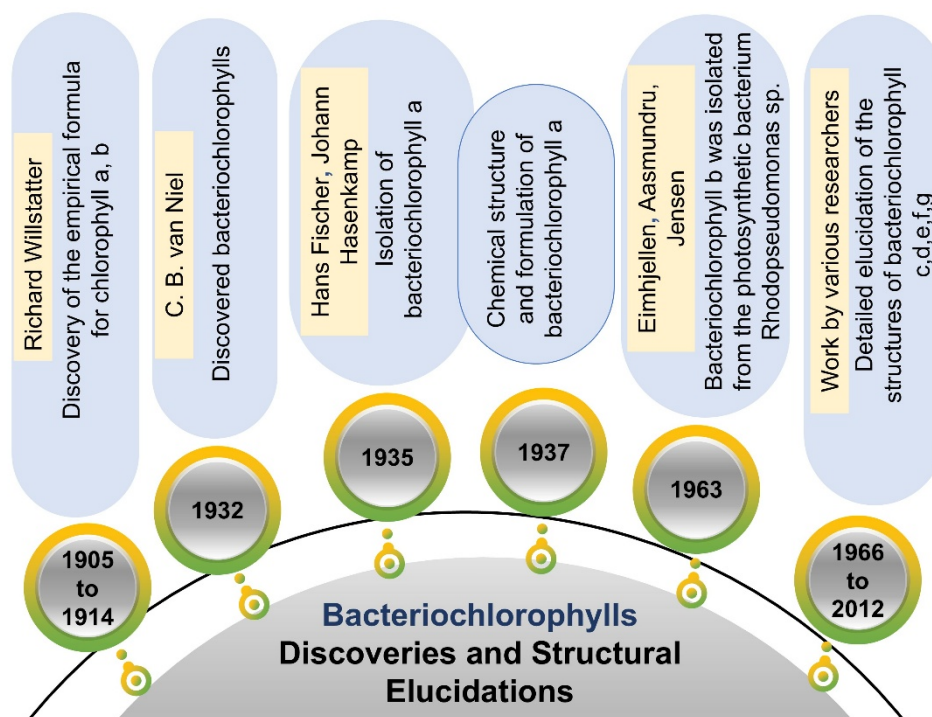


Figure 1: Timeline of Bacteriochlorophyll discoveries and structural elucidations.

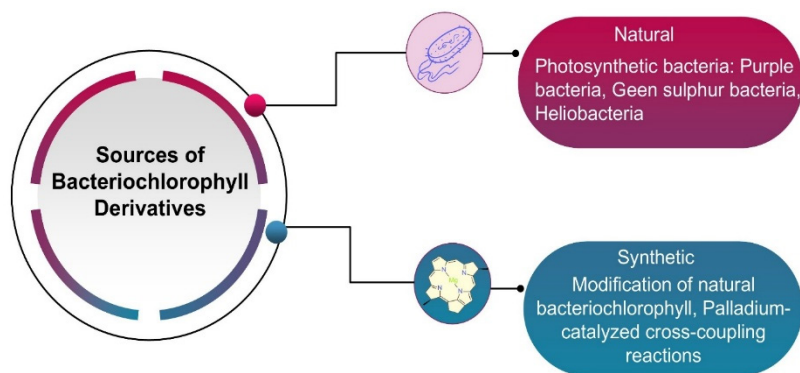


Figure 2: Sources of Bacteriochlorophyll derivatives.

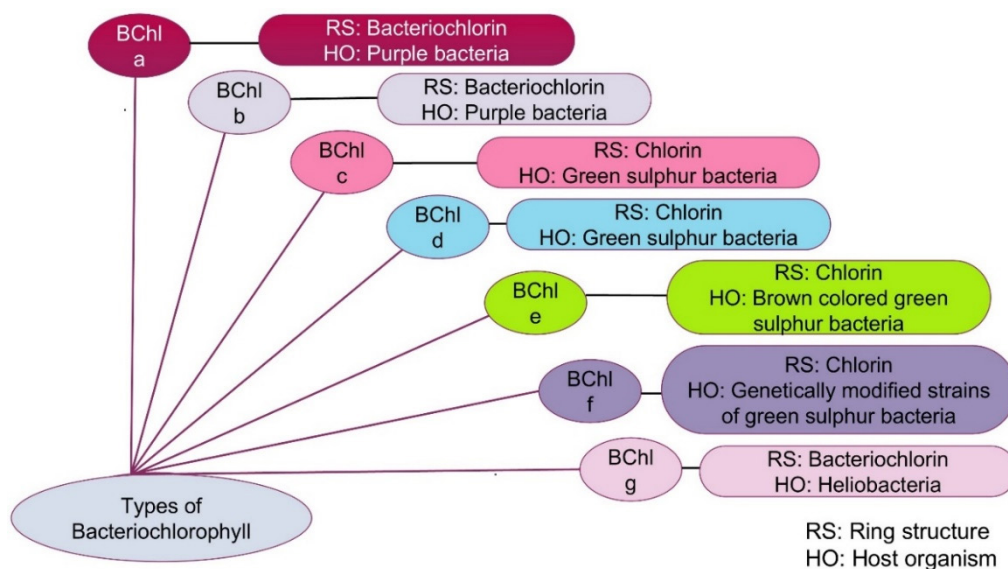


Figure 3: Types of Bacteriochlorophylls.

enabling controlled drug release.³⁰ Metallobacteriochlorophylls showed dual effects in PDT and chemotherapy.³¹ Palladium bacteriopheophorbide derivatives (Tookad® and Stakel®/Padeliporfin) are accepted for the treatment of localized prostate cancer employed in vascular targeted photochemotherapy. Stakel®, a photosensitizing derivative containing padeliporfin di-potassium, is activated upon absorption of light at NIR, thereby promoting the production of reactive oxygen radicals that ultimately damage the tumor cells.³² In research conducted by Goldshaid *et al.* (2010), the PDT potential and tumor imaging effects of STL-6009, a BChl d derivative, were determined. The study concluded that combining non-covalent binding to serum albumin and covalent association to cyclic RGDfK peptide effectively targets breast tumors, leading to more accumulation and prolonged retention.⁸² The detailed information highlighting the therapeutic relevance of bacteriochlorophyll derivatives has been represented in Figure 4 and Table 1.

Bacteriochlorophyll-c-stearyl

BChl-c-stearyl, which is a derivative of BChl c,⁸³ represented by formula $C_{52}H_{72}MgN_4O_4$ is a photosynthetic pigment usually

present in green photosynthetic bacteria.⁸⁴⁻⁸⁷ This tetrapyrrole derivative is distinguished by its stearyl ester chain at the C-17 propionate position.^{88,89} This side chain has a crucial role in light harvesting process involving chlorosomes.⁸³ BChl-c-stearyl related conjugate bacteriochlorophyll Cs has a long, straight-chain stearyl alcohol (Figure 5) contributing towards its distinct structural and functional characteristics.⁴⁷ BChl c stearyl is usually present in phototrophic bacteria,⁹⁰ but its existence in several plant extracts cannot be denied, as evidenced by the literature. It has been detected via GC-MS in extracts of several plants, as represented in Table 2, viz. *Byttneria herbacea*,⁸⁵ *Brassica oleracea*,⁹¹ *Lagenaria siceraria*,⁹² *Spermatidictyon suaveolens*,⁹³ *Madhuca longifolia*,⁹⁴ *Chenopodium album*,⁹⁵ *Rhodiola imbricata*,⁹⁶ *Ficus amplocarpa*,⁹⁷ *Kalanchoe pinnata*,⁹⁸ *Piper wightii*,⁹⁸ *Symplocos cochinchinensis*,⁹⁹ *Caryota urens*.¹⁰⁰ However, the presence of BChl-c-stearyl in plants is still under the point of discussion and research, as majorly they are identified as bacterial sources. Beyond bacterial sources, one derivative of bacteriochlorophyll c, specifically bacteriochlorophyll-c-stearyl, has been reported in several plant extracts. Although the function of bacteriochlorophyll-c-stearyl is not yet understood,

Table 1: Therapeutic applications of Bacteriochlorophyll derivatives.

Bacteriochlorophyll derivative	Source	Therapy involved	Targeted area	Outcome of the study	Reference
BChl-a	Rhodobacter sphaeroides	Antioxidant	Anti-aging	BChl-a promotes procollagen production and demonstrates significant anti-aging potential.	72
BChl-a	Photosynthetic bacteria	PDT	Ophthalmology	Management of corneal and scleral anomalies in ophthalmology.	73-75
BChl-a	Synthetic	Sonodynamic	LLC cells	BChl-a-mediated SDT increased cytotoxicity in LLC cells.	76
BChl-a	Reaction between bacteriopheophorbide methyl ether and diaminoalkanes	PDT	HEp2, A549, S37	At respective doses (2.5, 5.0 mg/kg), Bchl-a derivatives resulted in inhibition of tumor growth.	11
BChl	Rhodobacter sphaeroides	Anti-inflammatory	Cosmetic formulation	BChl, containing extract, displayed notable skin-protectant and moisturizing effects.	77
Photobac	BChl a	PDT	Glioblastoma	Due to its effectiveness in glioblastoma, with limited toxicity received approval for Phase I human clinical trials.	78
F2BMet (LUZ11 / Redaporfin [®])	BChl a	PDT	Head and neck tumors	Currently under phase II clinical trials for head and neck-associated tumors.	79,80
BChl-T	Rhodopseudomonas faecalis	PDT	Breast cancer cell (MCF-7) line	BChl-T demonstrated strong PDT potential against selected cell lines.	28
BChl-ser	Modified BChl-a derivative (C-17 propionic residue) conjugated with serine	PDT	M2R mouse melanoma cells	BChl-ser-treated tumors resulted in total oxygen depletion within the necrotic tissue.	29
BChl-ser	Photosynthetic bacteria	PDT	<i>In vivo</i> (animal tumour models)	BChl-ser combined with hyperthermia demonstrated tumour growth inhibition.	81
Bacteriochlorins (Tetrapyrroles)	BChla	PDT	Tumor	Bacteriochlorin nanoformulation improves tumor targeting.	30
Metallobacteriochlorophyll	BChl containing metallic ions: Pd, Zn, Ni, Co, Ru, Zn, Pt, Rh	PDT	Tumor	Ru, Pt, Pd, Rh provide cytotoxic effects; possibility for combined PDT and MBT.	31
Tookad [®] , Stakel [®]	Palladium bacteriopheophorbide	VTP	Targeted tumor destruction for prostate cancer	VTP improves the quality of life while managing low-risk localized prostate cancer.	32

Bacteriochlorophyll derivative	Source	Therapy involved	Targeted area	Outcome of the study	Reference
STL-6009	BChl d	PDT	Breast tumors	A combination of non-covalent binding to serum albumin and covalent association to c(RGDfK) enabled tumor target accumulation and increased retention of BChl derivatives in selected cancer (breast tumor).	82

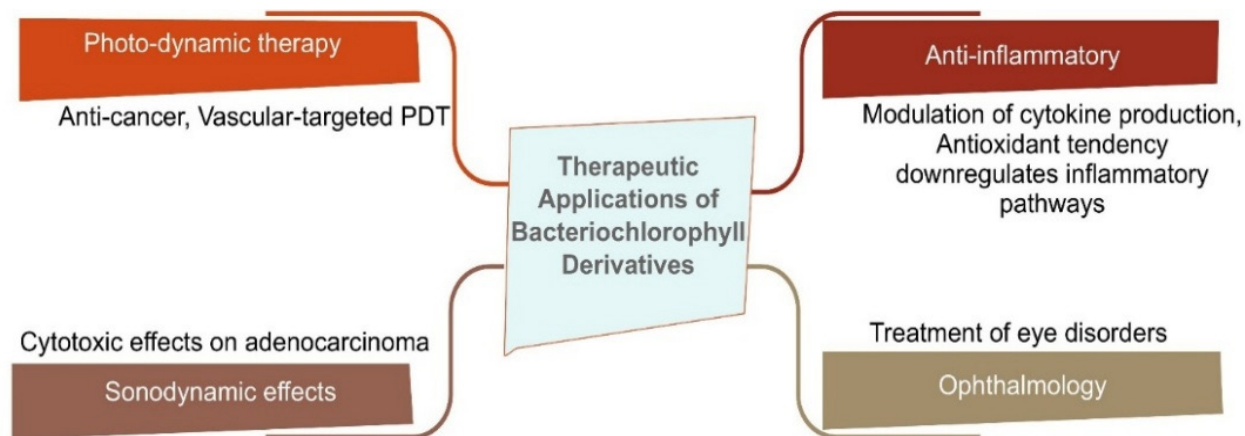


Figure 4: Therapeutic applications of Bacteriochlorophyll derivatives.

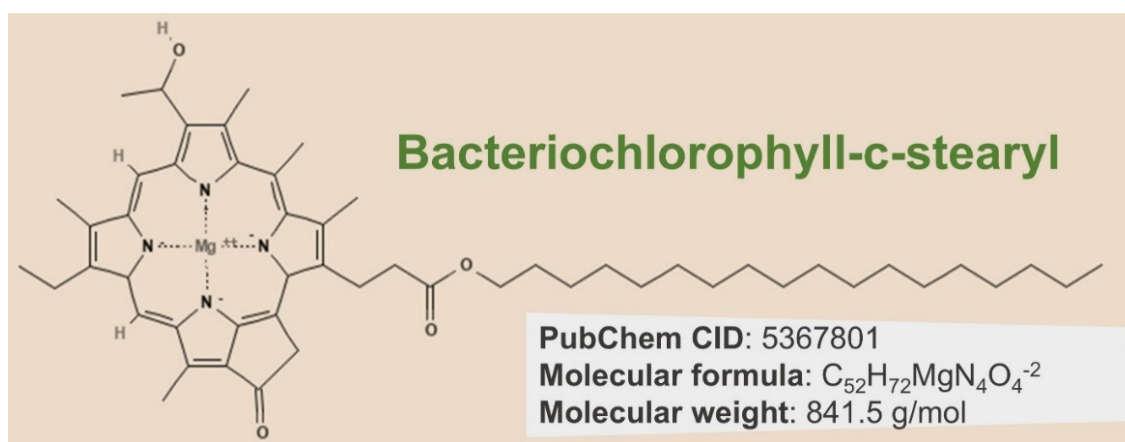


Figure 5: Structure of Bacteriochlorophyll-c-stearyl.

its intriguing detection in plant extracts opens a new line of discussion in plant biology.

Key Points and Challenges

Intrinsic (modifications in chemical structures, side chain and central atom substitutions) and extrinsic factors (light, temperature, pH, oxygen availability, solvent interactions) alter the stability of BChl derivatives to a greater extent. Hydrophilic BChl derivatives, viz. WST11 (TOOKAD) often face challenges

of less accumulation in tumor tissue and a rapid clearance rate due to non-covalent associations with serum albumin.¹⁰¹⁻¹⁰³ The presence of two reduced pyrrole rings in the chemical structure of BChl is attributed to its decreased stability.^{30,104} High amounts of purified natural BChl derivatives are required, resulting in high cost.^{52,105} Altered conjugation of BChl with cell-specific ligands, viz., tumor-specific antibodies, proteins, hormones, etc., is the major challenge in their usage as anti-tumor agents.^{73,106}

Table 2: Bacteriochlorophyll-c-stearyl reported in plants.

Plant	Peak % reported in GC-MS	Reference
<i>Rhodiola imbricata</i> , Crassulaceae	2.11	96
<i>Symplocos cochinchinensis</i> , Symplocaceae	0.81	99
<i>Spermadictyon suaveolens</i> , Rubiaceae	-	93
<i>Ficus amplocarpa</i> , Moraceae	1.95	97
<i>Byttneria herbaceae</i> , Sterculiaceae	3.84	85
<i>Chenopodium album</i> , Amaranthaceae	-	95
<i>Caryota urens</i> , Arecaceae	0.76	100
<i>Piper wightii</i> Piperaceae	1.32	98
<i>Lagenaria siceraria</i> , Cucurbitaceae	0.37	92
<i>Madhuca longifolia</i> , Sapotaceae	0.11	94
<i>Kalanchoe pinnata</i> Crassulaceae	1.71	86
<i>Brassica oleracea</i> , Brassicaceae	3.71	91

CONCLUSION

Bacteriochlorophyll derivatives possess significant therapeutic applications in ophthalmology, cosmetics, and as anti-inflammatory agents. Their unique ability for deep tissue penetration opens new diagnostic and therapeutic avenues, particularly for cancer and antimicrobial treatments. Bacteriochlorophyll derivatives have evolved into a highly versatile class of natural photosensitizers, showing immense promise for advancing photodynamic therapy and precision oncology. Overall, this review provides insight into the therapeutic applications of bacteriochlorophyll derivatives, emphasizing their usage in cancer treatment and hinting at novel roles yet to be completely understood in other biological contexts.

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None.

ABBREVIATIONS

BChl: Bacteriochlorophyll; **BChl a**: Bacteriochlorophyll a; **BChl b**: Bacteriochlorophyll b; **BChl c**: Bacteriochlorophyll c; **BChl d**: Bacteriochlorophyll d; **BChl e**: Bacteriochlorophyll e; **BChl f**: Bacteriochlorophyll f; **BChl g**: Bacteriochlorophyll g; **BChlide**: Bacteriochlorophyllide; **BChl-ser**: Bacteriochlorophyll-Serine; **BChl-T**: Bacteriochlorin-Trizma; **GC-MS**: Gas Chromatography-Mass Spectrometry; **HFF-1 cells**: Human

Foreskin Fibroblast Cells; **LLC cells**: Lewis Lung Carcinoma Cells; **M2R**: Mouse Melanoma Cell Line; **MBT**: Mentalization-Based Treatment; **MCF-7 cells**: Breast Cancer Cell Line; **NIR**: Near-Infrared; **PDT**: Photodynamic Therapy; **ROS**: Reactive Oxygen Species; **SDT**: Sonodynamic Therapy; **Tookad®**: Drug Used in Low-Risk Prostate Cancer; **Trizma®**: Tromethamine; U87, F98; **C6 cell lines**: Cancer Cell Lines Used in Glioblastoma Research; **VTP**: Vascular-Targeted Photodynamic Therapy.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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