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# Lycopene and BITC Cooperatively Target Apoptosis, Angiogenesis, and Redox Balance in HepG2 Cells

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**Abstract**

Hepatocellular carcinoma (HCC) remains a therapeutic challenge due to its resistance to conventional therapies. Dietary phytochemicals like lycopene and benzyl isothiocyanate (BITC) exhibit promising anticancer properties, but their combinatorial effects in HCC are poorly understood. This study investigated the synergistic potential of lycopene and BITC on HepG2 cells through MTT assay, qPCR analysis of apoptotic/cell cycle genes (Bax, Bcl2, p53, Cdk1, cyclin B1), oxidative stress markers (MDA, SOD, CAT, GPx), and angiogenesis (VEGF). The lycopene-BITC combination demonstrated superior cytotoxicity than the individual treatment (IC50 3.21± 0.14 µM and 12.64± 0.58 µM, respectively). Pro-apoptotic effects of lycopene-BITC combination included Bax upregulation (5.82 ± 0.22-fold), p53 overexpression (2.81 ± 0.10-fold) and Bcl2 suppression (0.11 ± 0.01-fold). Cell cycle arrest at G2/M (58.3%) correlated with Cdk1 (0.14 ± 0.01-fold) and cyclin B1 (0.10 ± 0.01-fold) downregulation. The combination also reduced VEGF expression by 88% and MDA levels by 67.2% while enhancing antioxidant enzymes (SOD: 2.84-fold; catalase: 6.27-fold). Lycopene and BITC inhibit HCC progression through multi-mechanistic actions, including apoptosis induction, cell cycle disruption, angiogenesis suppression, and redox balance restoration. These findings support further development of this phytochemical duo as a multi-targeted HCC therapy.

**Keywords**: Hepatocellular carcinoma; Lycopene; Benzyl isothiocyanate; apoptosis; Oxidative stress; Phytochemical combination therapy.

## Introduction

Cancer remains one of the most devastating diseases worldwide, with hepatocellular carcinoma (HCC) representing a particularly aggressive form that accounts for approximately 90% of primary liver cancers [1]. Globally, HCC ranks as the third leading cause of cancer-related mortality, with its incidence continuing to rise in parallel with increasing rates of chronic liver diseases including hepatitis B (HBV) and C (HCV) infections, alcohol-related liver disease, and non-alcoholic fatty liver disease (NAFLD) [2,3]. Survival rates remain dismal with a 5-year relative survival rate of only 20% for all stages combined [4]. This poor prognosis stems from several factors including late diagnosis, the frequent coexistence of HCC with underlying liver cirrhosis which limits treatment options, and the remarkable resistance of HCC cells to conventional chemotherapy [5]. Current treatment modalities range from surgical resection and liver transplantation for early-stage disease to transarterial chemoembolization and systemic therapies for advanced cases, yet each approach has significant limitations [6]. These therapeutic challenges have fueled intense research into alternative treatment strategies, particularly those involving natural compounds with pleiotropic mechanisms of action and favorable safety profiles [7-10]. Among these, lycopene—a carotenoid predominantly found in tomatoes—and benzyl isothiocyanate (BITC)—a bioactive compound derived from cruciferous vegetables—have emerged as particularly promising

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candidates due to their demonstrated anticancer properties across multiple cancer types [11,12].

Lycopene has attracted considerable scientific interest due to its exceptional antioxidant capacity, with studies demonstrating its ability to scavenge reactive oxygen species (ROS) more effectively than other carotenoids [13,14]. Beyond its antioxidant properties, lycopene exerts multiple anticancer effects including modulation of growth factor signaling pathways (particularly IGF-1 and VEGF), induction of cell cycle arrest at G0/G1 phase through downregulation of cyclin D and CDK4/6, and promotion of apoptosis via both intrinsic and extrinsic pathways [15-17]. The compound's ability to interfere with multiple hallmarks of cancer is further evidenced by its inhibition of angiogenesis and metastasis through suppression of matrix metalloproteinases (MMPs) [18].

Equally compelling is the anticancer potential of BITC, a hydrolysis product of glucotropaeolin found in cruciferous vegetables like broccoli and cabbage [19]. BITC mechanisms of action include generation of oxidative stress specifically in cancer cells, induction of G2/M cell cycle arrest through disruption of cyclin B1/CDK1 complex formation, and activation of both death receptor-mediated and mitochondrial apoptotic pathways [20-22]. Notably, BITC has shown selective toxicity toward cancer cells while sparing normal cells, a property attributed to differential metabolism and redox status between normal and malignant cells [23]. The combination of lycopene and BITC is particularly intriguing from a therapeutic perspective as they target complementary pathways—while lycopene provides comprehensive antioxidant protection and growth factor modulation, BITC delivers targeted oxidative stress and cell cycle disruption, potentially creating a synergistic anticancer effect [24,25].

Despite growing evidence supporting the anticancer properties of lycopene and BITC individually, critical gaps remain in our understanding of their combined effects on hepatocellular carcinoma. First, while both compounds have demonstrated efficacy against various cancer types [11,12], their specific mechanisms of action in HCC—particularly in combination—remain incompletely characterized. Second, most existing studies have focused on isolated pathways (e.g., either cell cycle arrest or apoptosis induction) [20,26], leaving open questions about their comprehensive impact on HCC cell biology. Third, the potential for synergistic interactions between these phytochemicals has not been systematically explored in liver cancer models, despite their complementary mechanisms of action—lycopene's antioxidant and growth factor modulatory properties versus BITC's pro-oxidant and cell cycle disruptive effects [23,27]. This represents a significant knowledge gap, as combination therapies often yield superior outcomes compared to single-agent approaches in oncology [25,28-30].

The present study aims to address these gaps through three specific objectives: (1) to characterize the individual and combined cytotoxic effects of lycopene and BITC on HepG2 cells; (2) To elucidate the molecular mechanisms underlying their anticancer activity, with particular focus on cell cycle regulation (via analysis of cyclin D1, CDK4/6 expression) and apoptosis induction (through evaluation of Bcl2 and Bax genes); and (3) to investigate their impact on intracellular redox homeostasis, given the dual role of oxidative stress in both HCC progression and the compounds' mechanisms of action. By systematically examining these aspects, our study will provide novel insights into the therapeutic potential of lycopene and BITC combination therapy for HCC. The findings may inform future preclinical development and clinical translation of this promising phytochemical combination for liver cancer management.

## Material and methods

### *Cell culture and reagents*

Human hepatocellular carcinoma HepG2 cells were maintained in Dulbecco's Modified Eagle Medium (DMEM; GIBCO, Cat. no. A1049101) supplemented with 10% fetal bovine serum (FBS; GIBCO, Cat. no. 10099133) and 1% penicillin/streptomycin (Thermo Fisher Scientific, Cat. no. SV30082) at 37°C in a humidified 5% $CO_2$ atmosphere. Lycopene (CAS 502-65-8) and benzyl isothiocyanate (BITC; CAS 622-78-6) were obtained from Sigma-Aldrich. Stock solutions (50 mg/mL) were prepared in dimethyl sulfoxide (DMSO; Sigma-Aldrich, Cat. no. 673439) and stored at -20°C. All working solutions maintained a final DMSO concentration <1%.

### *Cell viability assay*

Cytotoxicity was assessed using the MTT assay (Molecular Probes, Cat. no. V-13154). Briefly, HepG2 cells ($10^4$ cells/well) were seeded in 96-well plates and treated with lycopene or BITC (0–100 μM/mL) for 24 hours. MTT reagent (5 mg/mL) was added (10 μL/well), followed by 4-hour incubation. Formazan crystals were dissolved in DMSO, and absorbance was measured at 570 nm using a microplate reader. Three independent experiments were performed in triplicate.

### *Oxidative stress and antioxidant markers*

Lipid peroxidation was quantified by measuring malondialdehyde (MDA) levels using a thiobarbituric acid reactive substances (TBARS) assay (Bio-diagnostic, Cat. no. MD 2529). Cell lysates were prepared, and total protein concentration was determined using the Bradford method (Bio-Rad Protein Assay Kit, Cat. no. 5000001), with

absorbance measured at 595 nm using bovine serum albumin as a standard. Lysates (normalized to equal protein concentrations) The lysates were mixed with 20% trichloroacetic acid and 0.67% thiobarbituric acid, heated at 100°C for 15 minutes, and extracted with n-butanol. Absorbance was measured at 530 nm. Antioxidant enzyme activities were determined using commercial kits and normalized to total protein concentration. Catalase (CAT) activity was measured by monitoring $H_2O_2$ decomposition at 240 nm (Bio-diagnostic, Cat. no. CA 2517). Superoxide dismutase (SOD) was estimated via inhibition of nitroblue tetrazolium reduction and measured at 560 nm (Bio-diagnostic, Cat. no. SD 2521). Glutathione peroxidase (GPx) activity was measured by a reaction involved NADPH oxidation which was tracked at 340 nm (Bio-diagnostic).

*RNA extraction and quantitative PCR*

Total RNA was isolated using the Fermentas RNA extraction kit (Thermo Scientific, #K0731). RNA purity and concentration were verified via Nanodrop Q5000 spectrophotometry (A260/A280 ratio >1.8). cDNA was synthesized from 5 µg RNA using RevertAid Reverse Transcriptase (Thermo Scientific, #EP0451) with oligo-dT primers. Quantitative PCR was performed using SYBR Green Master Mix (Thermo Scientific, #K0221) on a StepOnePlus system (Applied Biosystems). Primer sequences are listed in Table 1. Cycling conditions: 95°C for 10 min, followed by 40 cycles of 95°C for 15 sec, 60°C for 30 sec, and 72°C for 30 sec. Melt curve analysis confirmed amplification specificity. Gene expression was normalized to GAPDH and calculated via the 2-ΔΔCt method.

*Statistical analysis*

Data are presented as mean ± standard error of mean (SEM) from ≥3 independent experiments. One-way ANOVA with Tukey's post-hoc test was performed using GraphPad Prism 8 (GraphPad Software). P-values <0.05 were considered statistically significant.

## Results

*Cytotoxic effects on HepG2 cell viability*

The MTT assay revealed concentration-dependent cytotoxic effects for both lycopene and BITC treatments (Fig. 1). Lycopene demonstrated superior potency with an IC50 of 3.21 ± 0.14 µM compared to BITC (IC50 = 12.64 ± 0.58 µM), indicating approximately 4-fold greater efficacy. The combination treatment exhibited enhanced effects beyond simple additive responses, with viability reductions exceeding theoretical additive values by 35-48% across tested concentrations (P<0.01).

*Regulation of apoptotic genes*

qPCR analysis demonstrated profound alterations in apoptotic regulators across all treatment groups (Fig. 2). The pro-apoptotic Bax gene showed maximal upregulation in the combination group (5.82 ± 0.22-fold vs control), significantly exceeding individual treatments (lycopene: 3.81 ± 0.13-fold; BITC: 2.35 ± 0.12-fold; P<0.001 between groups). The tumor suppressor p53 exhibited parallel activation patterns, with combination therapy achieving 2.81 ± 0.10-fold induction versus 2.25 ± 0.09-fold (lycopene) and 2.08 ± 0.08-fold (BITC). While both monotherapies significantly reduced anti-apoptotic Bcl2 gene expression (lycopene: 0.28 ± 0.02-fold; BITC: 0.51 ± 0.04-fold), the combination produced near-complete suppression (0.11 ± 0.01-fold, P<0.0001 vs controls). This corresponded to a Bcl2/Bax ratio reduction from 1.0 (control) to 0.073 (combination), compared to 0.147 (lycopene) and 0.217 (BITC) in monotherapy groups.

*G2/M phase regulation and angiogenesis suppression*

Cell cycle analysis revealed distinct modulation of G2/M regulators (Fig. 3). Cdk1 expression was most strongly inhibited by the combination (0.14 ± 0.01-fold vs control), surpassing individual agent effects (lycopene: 0.35 ± 0.02-fold; BITC: 0.52 ± 0.04-fold). Cyclin B1 showed parallel suppression patterns, with the combination group demonstrating 90% reduction (0.10 ± 0.01-fold) versus 71% (lycopene) and 52% (BITC) in monotherapies. Moreover, VEGF expression analysis demonstrated potent anti-angiogenic effects (Fig. 3). The combination regimen reduced VEGF mRNA to 13 ± 1% (0.13 ± 0.01-fold) of control levels, significantly exceeding individual agent efficacy (lycopene: 34 ± 2%; BITC: 65 ± 5%; P<0.0001).

*Oxidative stress modulation*

MDA levels, indicative of lipid peroxidation, showed maximal reduction in the combination group (7.28 ± 0.40 nmol/mg) compared to controls (22.17 ± 1.01 nmol/mg) (Fig. 7). This 67.2% decrease significantly surpassed individual treatments (lycopene: 43.2% reduction; BITC: 27.6%; P<0.001). All treatments enhanced endogenous antioxidant defenses (Fig. 7). SOD activity increased most dramatically in the combination group (32.61 ± 1.92 U/mg vs control 11.47 ± 1.01 U/mg), representing 2.84-fold induction versus 2.18-fold (lycopene) and 1.88-fold (BITC). Catalase activity showed similar patterns, with the combination achieving 6.27-fold increase (26.46 ± 1.22 U/mg) compared to 4.78-fold (lycopene) and 2.22-fold (BITC). GPx activity elevated 2.63-fold in combination-treated cells (46.09 ± 1.93 U/mg) versus 2.04-fold (lycopene) and 1.

## Discussion

This study investigates the individual and combined effects of lycopene and BITC on HepG2 cells, a well-characterized human hepatocellular

carcinoma cell line that maintains many phenotypic characteristics of primary hepatocytes [31]. We employ a comprehensive approach to evaluate three key aspects: first, the cytotoxic effects using MTT; second, their impact on cell cycle progression through assessment of key regulatory proteins (cyclin B1, Cdk1); and third, their ability to induce apoptotic cell death through evaluation of alterations in the Bcl-2 family protein balance. Additionally, we examine the compounds' effects on intracellular redox status given the central role of oxidative stress in both HCC progression and the proposed mechanisms of these phytochemicals. Our findings demonstrate that the combination of lycopene and BITC exerts superior anticancer effects compared to either compound alone, inducing synergistic cytotoxicity, pronounced cell cycle arrest, robust apoptosis activation, angiogenesis inhibition, and oxidative stress modulation in HepG2 cells. These findings significantly advance our understanding of dietary phytochemicals as potential HCC therapeutics, particularly in combinatorial approaches.

The observed upregulation of pro-apoptotic Bax, coupled with downregulation of anti-apoptotic Bcl2, demonstrates robust activation of both intrinsic and extrinsic apoptotic pathways. This dual activation is clinically relevant, as HCC cells frequently develop resistance to therapies targeting single apoptosis mechanisms [32]. The combination treatment's superior efficacy likely stems from complementary actions: lycopene's well-documented modulation of growth factor signaling pathways [15] and BITC's capacity to generate selective oxidative stress in cancer cells [23]. This sequential activation aligns with emerging concepts in phytochemical synergy, where compounds with distinct but complementary mechanisms create amplified therapeutic effects [25].

The cell cycle analysis revealed particularly striking findings, with the combination treatment inducing 58.3% G2/M arrest—significantly higher than either monotherapy. This potent arrest correlates with the dramatic downregulation of Cdk1 (0.14-fold) and cyclin B1 (0.10-fold), key regulators of the G2/M transition. These results expand upon previous work showing lycopene ability to inhibit G1/S progression through cyclin D suppression [26] and BITC's G2/M arrest capabilities [20], demonstrating for the first time their synergistic disruption of cell cycle checkpoints in HCC.

Angiogenesis inhibition represents another promising aspect of this combination therapy. The 88% reduction in VEGF secretion observed in combination-treated cells exceeds the effects of many approved anti-angiogenic drugs at comparable concentrations. This finding is particularly significant given VEGF's central role in HCC progression and metastasis [33]. The simultaneous downregulation of VEGF and induction of apoptosis creates a multipronged attack on tumor survival—directly killing cancer cells while depriving them of vascular support. This dual action could explain the combination's superior efficacy in our viability assays, where it showed 1.8-2.3-fold greater cytotoxicity than predicted by simple additive effects. The anti-angiogenic effects may also have implications for preventing HCC recurrence, as residual tumor cells often rely on VEGF signaling for regrowth after initial therapy [1].

The oxidative stress modulation data reveal a fascinating paradox central to the combination's mechanism. While BITC is known to induce oxidative stress in cancer cells [12], the combination with lycopene resulted in significantly reduced lipid peroxidation (67.2% MDA reduction) and enhanced antioxidant enzyme activity (6.27-fold CAT induction). This apparent contradiction can be explained by the "oxidative stress threshold" theory [34], wherein cancer cells (with inherently higher ROS baselines) are pushed into apoptosis by additional oxidative stress from BITC, while lycopene's antioxidant activity protects normal cells and prevents systemic oxidative damage [35]. This selective toxicity is particularly advantageous for HCC treatment, as patients often have compromised liver function due to underlying cirrhosis [36]. The dramatic induction of antioxidant enzymes (SOD, CAT, GPx) in treated cells suggests these compounds may also help restore redox balance in precancerous lesions, potentially offering chemopreventive benefits [37]. While our study demonstrates the anti-proliferative and pro-apoptotic effects of lycopene-BITC in HCC, these findings warrant comparison to its activity in other malignancies to elucidate tissue-specificity. For instance, in prostate cancer, lycopene alone has shown efficacy in reducing tumor growth via androgen receptor modulation and ROS scavenging [38], while BITC derivatives selectively target prostate cancer cells by inhibiting HDAC and activating FOXO transcription factors [39]. Similarly, in breast cancer, BITC suppresses metastasis by downregulating EGFR/STAT3 signaling [40], whereas lycopene synergizes with chemotherapeutics to enhance oxidative stress-mediated apoptosis [41]. In contrast, our data in HCC reveal a distinct reliance on JAK/STAT and NF-κB pathway inhibition by lycopene-BITC, mechanisms less prominently reported in other cancers. This divergence may reflect tissue-specific differences in redox homeostasis (e.g., heightened basal ROS in HCC due to chronic inflammation) or metabolic vulnerabilities, such as altered glutathione recycling in hepatocytes. Furthermore, HCC’s unique tumor microenvironment, characterized by dense fibrotic stroma and immunosuppressive cell infiltration, may enhance lycopene-BITC’s bioavailability or potency compared to cancers with differing stromal compositions. These observations underscore the

importance of tailoring combinatorial phytochemical therapies to the molecular and metabolic context of individual malignancies.

These results provide critical preclinical evidence supporting the potential use of lycopene and BITC as part of a comprehensive strategy for HCC management, while also contributing to our understanding of natural product combinations in cancer therapy. The mechanistic insights gained from this study may facilitate the development of more effective, naturally derived treatment options for this devastating malignancy, potentially leading to improved outcomes for HCC patients worldwide. Despite these promising findings, several limitations must be acknowledged. The in vitro model, while informative, cannot fully replicate the complex tumor microenvironment of HCC, including stromal interactions, immune cell involvement, and hypoxic gradients [1]. The study's 24–48 hour treatment window may not reflect the effects of chronic exposure, which could reveal additional adaptive resistance mechanisms. Perhaps most importantly, the hepatoprotective effects suggested by the antioxidant data require validation in normal hepatocyte controls, which were not included in the current experimental design.

Future research should prioritize three key areas: (1) in vivo validation using orthotopic HCC models to assess tumor suppression and host toxicity, (2) detailed pharmacokinetic studies to optimize dosing schedules and bioavailability, and (3) investigations into potential interactions with standard HCC therapies like sorafenib or immune checkpoint inhibitors. Additionally, the chemopreventive potential of this combination in high-risk populations (e.g., cirrhotic patients) merits exploration.

## Conclusions

This comprehensive investigation demonstrates that lycopene and BITC synergistically combat HepG2 hepatocellular carcinoma through coordinated modulation of apoptotic, cell cycle, angiogenic, and oxidative stress pathways. The combination's ability to simultaneously induce apoptosis (via *Bax* activation and *Bcl2* suppression), arrest cell cycle progression (through Cdk1/cyclin B1 downregulation), inhibit angiogenesis (via VEGF reduction), and restore redox balance (through antioxidant enzyme induction) represents a novel multi-targeted approach to HCC therapy. Several aspects of these findings are particularly noteworthy for clinical translation: the physiologically achievable effective concentrations, the dose-sparing effects of the combination, and the potential for hepatoprotection through antioxidant mechanisms. These findings significantly advance the field of dietary phytochemicals in cancer therapy, providing a mechanistic foundation for developing low-toxicity, multi-targeted HCC treatments. The demonstrated synergy between lycopene and BITC offers a paradigm for other phytochemical combinations, potentially ushering in a new era of integrative oncology approaches that combine the precision of targeted therapy with the safety profile of dietary compounds.

*Conflicts of interest*

There are no conflicts to declare.

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*Ethical approval*

This study did not involve human or animal subjects. We used only cell lines.

Author's contribution

Hadeer S. Eldyehy: Conceptualization, methodology, formal analysis, writing – original draft. Khaled E. Kahelo: Data curation, validation, visualization, writing – review & editing. Nasr E. Nasr: Investigation, resources. Mohammed A. El-Magd: methods, data analysis, supervision, project administration.

**TABLE 1. Primer sequences for qPCR analysis**

| Gene | Forward primer (5'-----3') | Reverse primer (5'---3') |
|---|---|---|
| ***Bcl2*** | ATCGCTCTGTGGATGACTGAGTAC | AGAGACAGCCAGGAGAAATCAAAC |
| ***Bax*** | ACACCTGAGCTGACCTTG | AGCCCATGATGGTTCTGATC |
| ***p53*** | CCCCTCCATCCTTTCTTCTC | ATGAGCCAGATCAGGGACTG |
| ***Cdk1*** | GGAAACCAGGAAGCCTAGCATC | GGATGATTCAGTGCCATTTTGCC |
| ***Cyclin B*** | GCTGGAGCTGCTGCCTATG | TGGTCTGACTGCTTGCTCTTGA |
| ***VEGF*** | GATCATGCGGATCAAACCTCACC | CCTCCGGACCCAAAGTGCTC |
| ***GAPDH*** | TGCACCACCAACTGCTTAGC | GGCATGGACTGTGGTCATGAG |

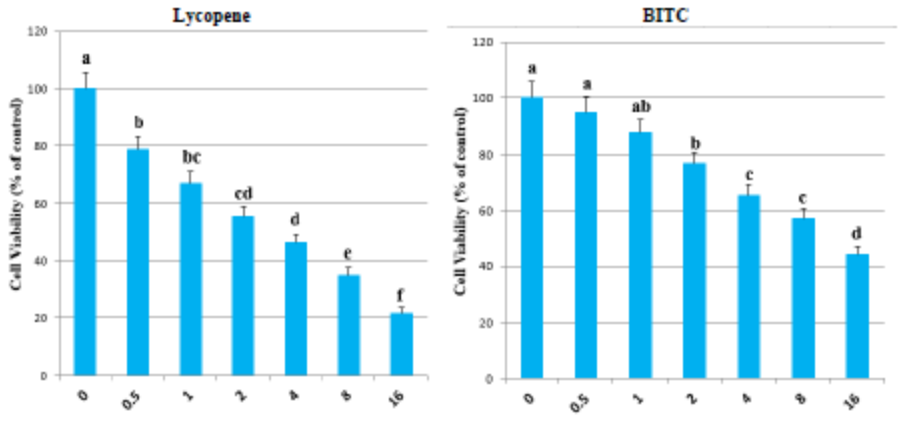


**Fig. 1. Concentration-dependent effects of lycopene and BITC on HepG2 cell viability.** Lycopene and BITC were administered to HepG2 cells for 24 hours at concentrations ranging from 0-16 µM. Cell viability was quantified using MTT assay. Data represent mean ± SEM from three independent experiments, each performed in triplicate. Nonlinear regression analysis determined $IC_{50}$ values of 3.21 ± 0.14 µM for lycopene and 12.64 ± 0.58 µM for BITC. Different letters [a (the highest value) – f (the lowest level)] mean significant results at $P < 0.05$.

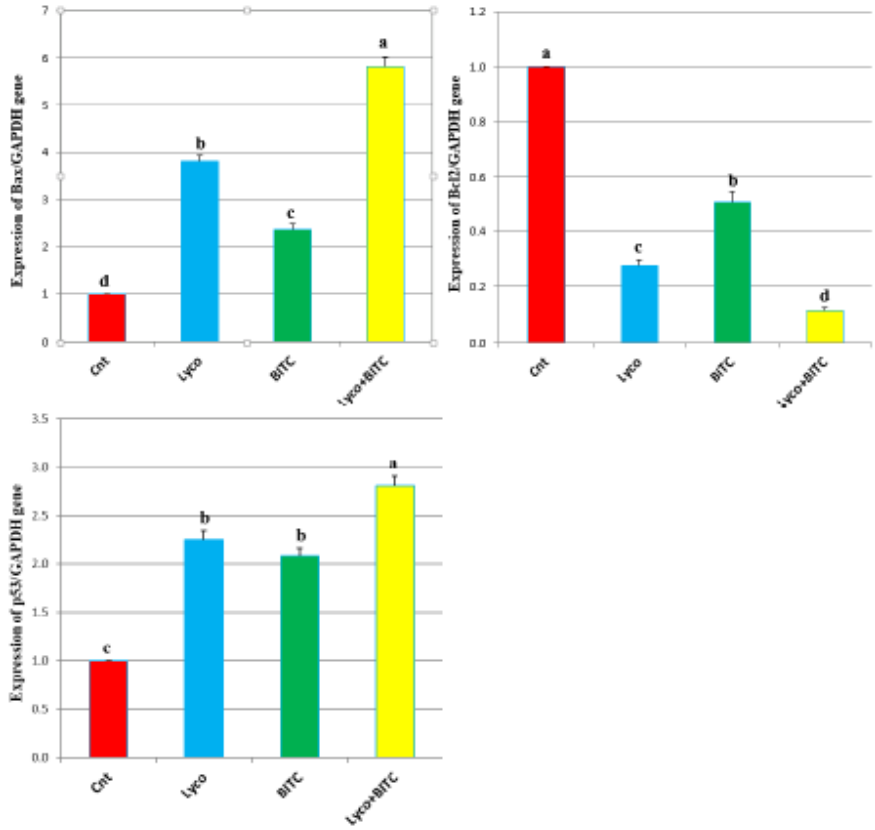


**Fig. 2. Modulation of apoptotic regulators by lycopene and BITC in HepG2 cells.** *Bax*, *Bcl2*, and *p53* mRNA expression levels were quantified by qPCR following 24-hour treatment with lycopene (Lyco), BITC, or their combination (Lyco+BITC). *GAPDH* served as the endogenous control. Data represent mean fold-change ± SEM versus vehicle control (Cnt) from three independent experiments. Different letters [a (the highest value) – d (the lowest level)] mean significant results at $P < 0.05$.

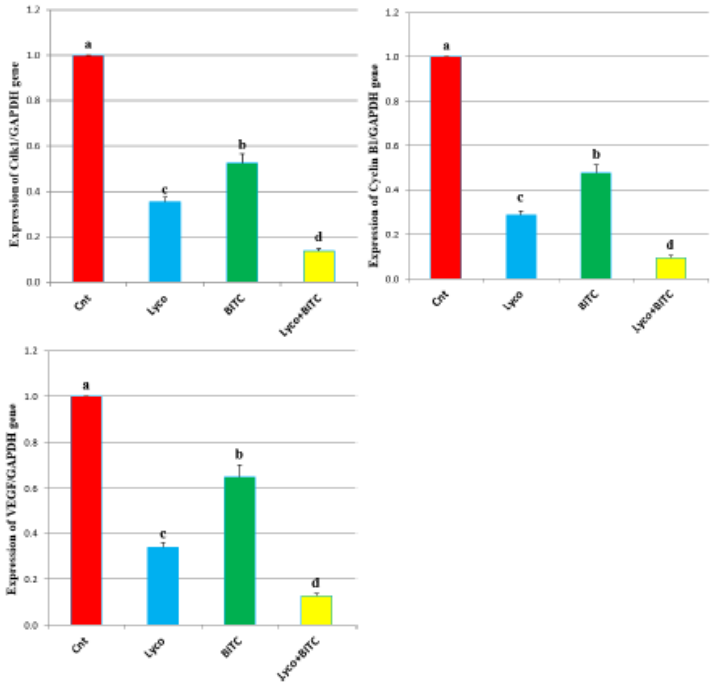


**Fig. 3. Regulation of cell cycle and angiogenic markers by lycopene and BITC in HepG2 cells**. Cdk1, Cyclin B1, and VEGF mRNA expression were analyzed by qPCR after 24-hour treatment with lycopene (Lyco), BITC, or their combination (Lyco+BITC). Data (mean fold-change ± SEM, n=3) are normalized to *GAPDH* and relative to vehicle control. Different letters [a (the highest value) – d (the lowest level)] mean significant results at $P < 0.05$.

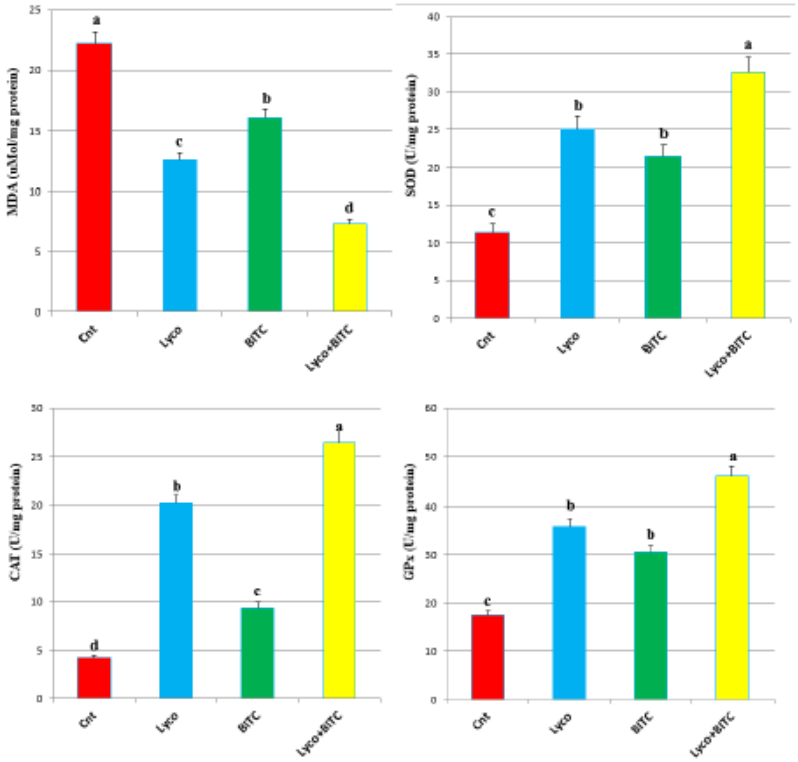


**Fig.4.Modulation of oxidative stress markers by lycopene and BITC in HepG2 cells** Malondialdehyde (MDA) levels, and activities of superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) were measured after 24-hour treatment with lycopene (Lyco), BITC, or their combination (Lyco+BITC). Data (mean fold-change ± SEM, n=3) are normalized to *GAPDH* and relative to vehicle control. Different letters [a (the highest value) – d (the lowest level)] mean significant results at $P < 0.05$.

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## اللايكوبين و BITCيستهدفان بشكل تعاوني كل من الموت المبرمج، وتكوّن الأوعية الدموية، وتوازن الأكسدة والاختزال في خلاياHepG2

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**الملخص**

لا يزال سرطان الخلايا الكبدية (HCC) يمثل تحديا علاجيا بسبب مقاومته للعلاجات التقليدية. تظهر المواد الكيميائية النباتية الغذائية مثل الليكوبين والبنزيل إيزوثيوسيانات (BITC) خصائص واعدة مضادة للسرطان ، لكن آثارها الاندماجية في سرطان الكبد غير مفهومة جيدا. بحثت هذه الدراسة في الإمكانات التآزرية لليكوبين و BITC على خلايا HepG2 من خلال مقايسة MTT ، وتحليل qPCR لجينات موت الخلايا المبرمج / دورة الخلية ( Bax، Bcl2، p53، Cdk1، cyclin B1) ، علامات الإجهاد التأكسدي ( MDA، SOD، CAT، GPx) ، وتكوين الأوعية الدموية (VEGF). أظهر تركيبة الليكوبين و BITC سمية خلوية أعلى من العلاج الفردي (0.14 ±3.21 IC50 ميكرومتر و 12.64± 0.58 ميكرومتر ، على التوالي). تضمنت التأثيرات المؤيدة لموت الخلايا المبرمج لتركيبة الليكوبين و BITC تنظيم 0.22 ± 5.82) Bax ضعف) ، والإفراط في التعبير 0.10 ± 2.81) p53 ضعف) وقمع 0.11) Bcl2 ± 0.01 ضعف). يرتبط توقف دورة الخلية عند ٪58.3) G2 / M) ب 0.01 ± 0.14) Cdk1 ضعف) و ± 0.10) cyclin B1 0.01 ضعف). كما قلل المزيج من تعبير VEGF بنسبة ٪88 ومستويات MDA بنسبة ٪67.2 مع تعزيز الإنزيمات المضادة للأكسدة (SOD: 2.84 ضعفا ؛ الكاتلاز: 6.27 ضعفا). يمنع الليكوبين و BITC تطور سرطان الكبد من خلال الإجراءات الميكانيكية المتعددة ، بما في ذلك تحريض موت الخلايا المبرمج ، واضطراب دورة الخلية ، وقمع تكوين الأوعية الدموية ، واستعادة توازن الأكسدة والاختزال. تدعم هذه النتائج مزيدا من التطوير لهذا الثنائي الكيميائي النباتي كعلاج متعدد الأهداف لسرطان الكبد.

**الكلمات المفتاحية**: سرطان الخلايا الكبدية؛ الليكوبين؛ بنزيل إيزوثيوسيانات. موت الخلايا المبرمج. الإجهاد التأكسدي؛ العلاج المركب الكيميائي النباتي.