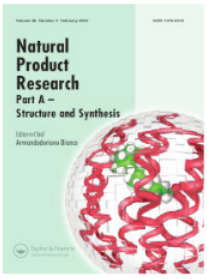
Bixin

by Paper Npr

Submission date: 11-May-2023 07:01PM (UTC+0700)
Submission ID: 2090334605
File name: Natural_Product_research.pdf (1.07M)
Word count: 3534
Character count: 19474



Natural Product Research
Formerly Natural Product Letters

ISSN: (Print) (Online) Journal homepage: https://www.tandfonline.com/loi/gnpl20

Bixa orellana L. carotenoids: antiproliferative activity on human lung cancer, breast cancer, and cervical cancer cells *in vitro*

Lia Kusmita, Yuvianti Dwi Franyoto, Mutmainah Mutmainah, Ika Puspitaningrum & Agustina D. R. Nurcahyanti

To cite this article: Lia Kusmita, Yuvianti Dwi Franyoto, Mutmainah Mutmainah, Ika Puspitaningrum & Agustina D. R. Nurcahyanti (2022): *Bixa orellana* L. carotenoids: antiproliferative activity on human lung cancer, breast cancer, and cervical cancer cells *in vitro*, Natural Product Research, DOI: 10.1080/14786419.2022.2036144

To link to this article: https://doi.org/10.1080/14786419.2022.2036144

NATURAL PRODUCT RESEARCH
https://doi.org/10.1080/14786419.2022.2036144

Taylor & Francis
Taylor & Francis Group

SHORT COMMUNICATION

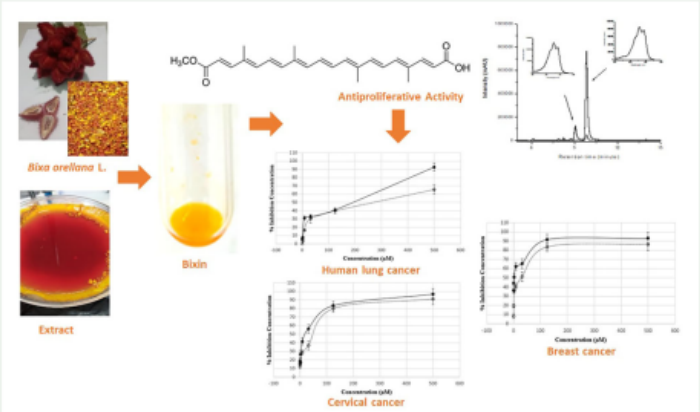
Bixa orellana L. carotenoids: antiproliferative activity on human lung cancer, breast cancer, and cervical cancer cells *in vitro*

Lia Kusmita[a], Yuvianti Dwi Franyoto[a], Mutmainah Mutmainah[a], Ika Puspitaningrum[a] and Agustina D. R. Nurcahyanti[b]

[a]Department of Pharmacy, STIFAR Yayasan Pharmasi Semarang, Plamongansari Pucanggading Semarang, Indonesia; [b]Department of Pharmacy, School of Medicine and Health Sciences, Atma Jaya Catholic University of Indonesia, Jakarta, Indonesia

ABSTRACT

Emerging evidence on the potential pro-oxidant effect of carotenoids provokes apoptosis of cancer cells. *Bixa orellana* L. is native to Central and South America, interestingly, is also cultivated worldwide. Apo-carotenoids present in *B. orellana* L. are mainly dominated by bixin and norbixin and demonstrate fundamental antioxidant activity. Anti-proliferative activity on human cancer cells is rarely investigated. We isolated bixin from *B. orellana* L. found in the island of Java using Ultra-Fast Liquid Chromatography and confirmed the isolated compound using Liquid Chromatography-MS/MS. Bixin and crude extract were examined on human lung cancer (A549), cervical cancer (HeLa), and breast cancer (MCF-7). Anti-proliferative activity revealed to be promising on both, the isolated pigment and crude extract. Further investigation on the mechanism of action and effect on other cell lines, both *in vitro* and *in vivo*, are required before clinical translation.



ARTICLE HISTORY
Received 28 September 2021
Accepted 24 January 2022

KEYWORDS
Bixa orellana L.; Bixaceae; bixin; annatto pigment; pro-oxidant; anti-proliferative; lung cancer; cervical cancer

CONTACT Agustina D. R. Nurcahyanti adr.nurcahyanti@atmajaya.ac.id

Supplemental data for this article can be accessed online at https://doi.org/10.1080/14786419.2022.2036144.

1. Introduction

Metabolic reaction in normal cells produces reactive oxygen species (ROS) by-products (Liguori et al. 2018). The intracellular antioxidant system scavenges ROS and prevents cellular damage. Antioxidant molecules, such those polyphenols, flavonoids, and member of carotenoids have been known for antioxidant effect and have furnished optimistic activity in a particular clinical setting (Wang et al. 2015; Song et al. 2017; Beydoun et al. 2019; Rattanavipanon et al. 2021). Diverse functional groups and isomer configuration of *cis-* (*-Z*) and *trans-* (*-E*) on the hydrocarbon backbone affect the bioactivity of carotenoids, in which the *Z*-isomers often demonstrate higher bioavailability and antioxidant activity, also often associated with thrilling pro-oxidant actions in lipid peroxidation (Shin et al. 2020). The bioactivity of carotenoids also depends on the concentration that can affect the presence of other carotenoids, oxygen, and new free radicals as observed in the study using MCF-7 breast cancer cells (Sowmya et al. 2017) and human leukemia HL-60 cells (Ganesan et al. 2011). Unlikely normal cells, cancer cells generate a higher amount of ROS due to the rapid metabolism of the cancerous cell, increased metabolism dysfunction, increased lipid peroxidation, and the failure of antioxidant enzymes (Liguori et al. 2018). Few studies have suggested the dual function of carotenoids as an antioxidant in normal cells and also pro-oxidant function induced apoptosis in cancer cells (Shin et al. 2020). Characteristics of cancer cells also determine the cytotoxicity activity of carotenoids, as seen in estrogen receptor (ER) negative MDA-MB-231 human breast cancer cells that are more susceptible to lycopene treatments than the ER-positive MCF-7 cells (Gloria et al. 2014). Understanding carotenoid's fate on the biology system, on how carotenoids turn pro-oxidant and antioxidant in specific types of cells and tissues, is warranted. One of the purposes is to optimize the anti-proliferative modulation on human cancer cells.

A well-known carotenoid producing species, *Bixa orellana* L., is a small shrubby tree native to Central and South America and used as traditional medicine (Giorgi et al. 2013). Between the 16th and 17th centuries, *B. orellana* L. was distributed to Southeast Asia, African, the Caribbean, and has since been cultivated in tropical regions including India, Srilanka, and Java (Ventosa 2018). Bixin constitutes 70–80% of all pigments present in *B. orellana* L. seed membrane while norbixin makes up the remaining 20% (Reith and Gielen 1971). These two pigments are also referred to as annatto and are traditionally used as natural food coloring (Bouvier et al. 2003; Raddatz-Mota et al. 2017) and cosmetic (Bouvier et al. 2003; Ntohogian et al. 2018). In the context of traditional medicine, some studies demonstrate broad pharmacological activities of *B. orellana* L. seeds, such as antioxidant activity, nephroprotective effect, and lipid-lowering effect (Kiokias and Gordon 2003; Paula et al. 2009; Rivera-Madrid et al. 2016; Souza et al. 2016; Roehrs et al. 2017).

Cancer chemopreventive compounds from natural products, including from the family of tocotrienols, is of interest due to the unique chemical structure resulting in broader pharmacological effect and reduced toxicity-induced side effects when compared to synthetic anticancer drugs (Marelli et al. 2019). Few studies on annatto exhibit selective antimyeloma activity (Tibodeau et al. 2010), induce apoptosis of human Hep3B hepatocellular carcinoma cells (Kumar et al. 2018), and tumor growth inhibition in HER-2/neu transgenic mice (Pierpaoli et al. 2013). Average daily intake of

annatto extract in North American and some Latin American countries is higher as compared to Acceptable Daily Intake (ADI) regulated by WHO and Committee of Food Additives (0–12 mg/kg body weight). No toxicity issue is found after administration of annatto by mammals at high levels (Hagiwara et al. 2003; Bautista et al. 2004). Potency of annatto as cancer chemopreventive effects is indeed promising. Cancer is a complex genetic disease. Extensive heterogeneity within patients' cancer cells has important implications on the type of treatment provided (Loeb et al. 2003). Thus, an in-depth investigation of annatto on various cell types of cancer is highly required, especially to measure its efficacy and ensure safe transition into the clinics.

We further investigated, for the first time, inhibition of cancer proliferation by annatto pigment and crude extract of *B. orellana* L. collected from Java (Figure S1) in three most prevalent cancer cell lines. Anti-proliferative effect and potency of pro-apoptosis was discussed to gain broader insight for further in-depth investigation.

2. Results and discussion

The average yield of seed extract was 3.73 ± 0.33%. Kurniawati et al. (2007) confirmed that yield of annatto seed extract is 3.75% (Kurniawati et al. 2010). Samples from previous and current study were sourced from the same area. We extended our current study to isolate two main carotenoids, bixin, and norbixin. Acetone was used to dilute the carotenoid phase gathered from the fractionation. The diluted carotenoid was identified using UFLC in which Bixin was detected at retention time t_R = 9.819 min and norbixin at retention time t_R= 9.046 min (Figure S2). Isolated bixin from column chromatography has been confirmed using LC-MS/MS. Fragmented spectra at 450 nm indicates the presence of bixin, with fragmentation 377 $[M+H\text{-}18]^+$, 363 $[M+H\text{-}32]^+$, 335 $[M+H\text{-}32\text{-}28]^+$, 317 *m/z* (Figure S6-9), similar with available references (Chisté et al. 2011). Norbixin was not further identified as only low amounts were obtained.

The current study investigated the anti-proliferative activity of the crude extract and isolated bixin that had not been performed in the previous study. Antiproliferative assay shows profound results on three types of cells, especially HeLa and MCF-7, with the crude extract having greater activity than the isolate bixin (Figure S10a–c). IC_{50} of crude extract were 185.54 ± 14.46, 17.90 ± 3.00, and 2.06 ± 0.30 μM, while IC_{50} of isolated bixin were 259.38 ± 28.98, 43.87 ± 3.06, and 14.38 ± 0.55 μM against A549, HeLa, and MCF-7 cells, respectively.

Cis-bixin has been identified to possess profound anti-myeloma effect (Tibodeau et al. 2010). *Cis-trans* isomerization of bixin was discovered in the early year of discovery (Zechmeister 1960). Bixin was confirmed as the apo-carotenoid 90-*cis*-bixin (methyl hydrogen 90-*cis*-6,60-diapocarotene-6,60-dioate, $C_{25}H_{30}O_4$), and commonly referred to as *cis*-bixin. *Cis* -bixin is normally soluble in most polar organic solvents showing an orange color (Scotter 2009). However, during exposure in solution, *cis*-bixin may be converted to the all-*trans* isomer due to its instability. *Trans*-bixin is the more stable isomer, has similar structure to the *cis*-isomer, but shows a red color in solution (Scotter 2009). During extraction and fractionation process, *cis*-bixin can possibly undergo isomerization, giving a mixture of all-*trans*- and *cis*-bixin in variable proportions and other degradation products, depend on the solvent used during extraction,

temperature, and time. The presence of *cis*-bixin in the extract form may prevent isomerization to all-*trans*-bixin, and thus possibly contribute to the greater activity than isolated bixin (Figure S10a–c).

A549 cells show reduced sensitivity to annatto seed extract and bixin when compared to other cell lines used in this study, HeLa and MCF-7 (Figure S10a–c). The reason underlying this reduced activity is most likely due to the mechanism of multidrug resistance reported in A549, including activation of efflux pump by drug transporter (Xu et al. 2014; Lin et al. 2020) and increased mitochondrial activity (Gao et al. 2019). Several clinical and pre-clinical studies support evidence that protein transporter plays a vital contribution to the multidrug resistance of cancer cells (Xiao et al. 2021), such as ABCC1 found in human small-cell lung cancer cell lines (Cole et al. 1992). The resistance of lung cancer A549 to crude extract and isolated bixin may be due to carotenoids acting as the substrate of drug transporter, which is overexpressed in the A549 cells. Carotenoids, as a substrate, bind to the transporter (Tinoush et al. 2020). However, without sufficient concentration, carotenoids were not able to enter cancer cells and interfere with intracellular processes, and thus resulting in failure to inhibit growth and/or promote apoptosis.

Likely, it has been shown against hepatocellular carcinoma (Kumar et al. 2018), myeloma cells (Tibodeau et al. 2010), melanoma cells (Anantharaman et al. 2016; de Oliveira Júnior et al. 2019), and human leukemia cell (Santos et al. 2016). One proposed mechanism underlying this activity is via ROS-mediated apoptosis (Tibodeau et al. 2010; Anantharaman et al. 2016; Santos et al. 2016; de Oliveira Júnior et al. 2019). Carotenoids are a popular antioxidant agent with the ability to modulate the intracellular redox status, exerting both antioxidant and pro-oxidant properties, depending on concentration, cell types, and microenvironment of the cells (Sznarkowska et al. 2017). Besides the modulation on drug transporter, the mechanism underlying the unfavorable interaction between annatto seed extract and isolated bixin in A549 cells can also be due to the redox status, in which modulation of natural antioxidant activity was more significant than the capacity as pro-oxidant induced apoptosis.

3. Experimental

Provided in Supplementary File

4. Conclusions

Annatto seed extract performed profound anti-proliferative activity when compared to isolated bixin in three types of human cancer cell lines. Stable form of *cis*-bixin in the extract, along with the combination norbixin and other carotenoids, may exert more potent activity instead of the single isolated bixin. Redox signaling pathway-induced apoptosis and modulation on drug transporter requires further in-depth investigation on a specific type of cancer and tumor. It may generate rational cancer prevention and therapy including its combination with other conventional chemotherapeutic drugs.

Disclosure statement

Authors declare no conflict of interest.

Funding

This research was funded by the Indonesian Ministry of Research, Technology, and Higher Education, under Research Grant No.1208/LL3/PG/2021.

ORCID

Agustina D. R. Nurcahyanti http://orcid.org/0000-0002-5690-8742

References

Anantharaman A, Hemachandran H, Mohan S, Manikoth Ayyathan D, D TK, C GPD, Siva R. 2016. Induction of apoptosis by apocarotenoids in B16 melanoma cells through ROS-mediated mitochondrial-dependent pathway. J Funct Foods. 20(C):346–357.

Bautista ARPL, Moreira ELT, Batista MS, Miranda MS, Gomes ICS. 2004. Subacute toxicity assessment of annatto in rat. Food Chem Toxicol. 42(4):625–629.

Beydoun MA, Chen X, Jha K, Beydoun HA, Zonderman AB, Canas JA. 2019. Carotenoids, vitamin A, and their association with the metabolic syndrome: a systematic review and meta-analysis. Nutr Rev. 77(1):32–45.

Bouvier F, Dogbo O, Camara B. 2003. Biosynthesis of the food and cosmetic plant pigment bixin (annatto). Science. 300(5628):2089–2091.

Chisté RC, Yamashita F, Gozzo FC, Mercadante AZ. 2011. Simultaneous extraction and analysis by high performance liquid chromatography coupled to diode array and mass spectrometric detectors of bixin and phenolic compounds from annatto seeds. J Chromatogr A. 1218(1): 57–63..

Cole SP, Bhardwaj G, Gerlach JH, Mackie JE, Grant CE, Almquist KC, Stewart AJ, Kurz EU, Duncan AM, Deeley RG, et al. 1992. Overexpression of a transporter gene in a multidrug-resistant human lung cancer cell line. Science. 258(5088):1650–1654.

Ganesan P, Noda K, Manabe Y, Ohkubo T, Tanaka Y, Maoka T, Sugawara T, Hirata T. 2011. Siphonaxanthin, a marine carotenoid from green algae, effectively induces apoptosis in human leukemia (HL-60) cells. Biochim Biophys Acta. 1810(5):497–503.

Gao Y, Dorn P, Liu S, Deng H, Hall SRR, Peng R-W, Schmid RA, Marti TM. 2019. Cisplatin-resistant A549 non-small cell lung cancer cells can be identified by increased mitochondrial mass and are sensitive to pemetrexed treatment. Cancer Cell Int. 19(1):317–331.

Giorgi A, De Marinis P, Granelli G, Chiesa LM, Panseri S. 2013. Secondary metabolite profile, antioxidant capacity, and mosquito repellent activity of *Bixa orellana* from Brazilian Amazon region. J Chem. 2013:1–10.

Gloria NF, Soares N, Brand C, Oliveira FL, Borojevic R, Teodoro AJ. 2014. Lycopene and beta-carotene induce cell-cycle arrest and apoptosis in human breast cancer cell lines. Anticancer Res. 34(3):1377–1386.

Hagiwara A, Imai N, Doi Y, Nabae K, Hirota T, Yoshino H, Kawabe M, Tsushima Y, Aoki H, Yasuhara K, et al. 2003. Absence of liver tumor promoting effects of annatto extract (norbixin), a natural carotenoid food color, in a medium-term liver carcinogenesis bioassay using male F344 rats. Cancer Lett. 199(1):9–17.

de Oliveira Júnior RG, Bonnet A, Braconnier E, Groult H, Prunier G, Beaugeard L, Grougnet R, da Silva Almeida JRG, Ferraz CAA, Picot L, et al. 2019. Bixin, an apocarotenoid isolated from *Bixa*

orellana L., sensitizes human melanoma cells to dacarbazine-induced apoptosis through ROS-mediated cytotoxicity. Food Chem Toxicol. 125:549–561..

Kiokias S, Gordon MH. 2003. Antioxidant properties of annatto carotenoids. Food Chem. 83(4): 523–529.

Kumar Y, Phaniendra A, Periyasamy L. 2018. Bixin triggers apoptosis of human Hep3B hepatocellular carcinoma cells: an insight to molecular and in silico approach. Nutr Cancer. 70(6): 971–983.

Kurniawati PT, Soetjipto H, Limantara L. 2007. Antioxidant and antibacterial activities of bixin pigment from annatto (*Bixa orellana* L.) seeds. Indones J Chem. 7(1):88–92..

Liguori I, Russo G, Curcio F, Bulli G, Aran L, Della-Morte D, Gargiulo G, Testa G, Cacciatore F, Bonaduce D, et al. 2018. Oxidative stress, aging, and diseases. Clin Interv Aging. 13:757–772.

Lin H, Hu B, He X, Mao J, Wang Y, Wang J, Zhang T, Zheng J, Peng Y, Zhang F, et al. 2020. Overcoming taxol-resistance in A549 cells: A comprehensive strategy of targeting P-gp transporter, AKT/ERK pathways, and cytochrome P450 enzyme CYP1B1 by 4-hydroxyemodin. Biochem Pharmacol. 171:113733.

Loeb LA, Loeb KR, Anderson JP. 2003. Multiple mutations and cancer. Proc Natl Acad Sci USA. 100(3):776–781.

Marelli M, Marzagalli M, Fontana F, Raimondi M, Moretti RM, Limonta P. 2019. Anticancer properties of tocotrienols: a review of cellular mechanisms and molecular targets. J Cell Physiol. 234(2):1147–1164.

Noppe H, Abuin Martinez S, Verheyden K, Van Loco J, Companyo Beltran R, De Brabander HF. 2009. Determination of bixin and norbixin in meat using liquid chromatography and photodiode array detection. Food Addit Contam Part A Chem Anal Control Expo Risk Assess. 26(1): 17–24.

Ntohogian S, Gavriliadou V, Christodoulou E, Nanaki S, Lykidou S, Naidis P, Mischopoulou L, Barmpalexis P, Nikolaidis N, Bikiaris D, et al. 2018. Chitosan nanoparticles with encapsulated natural and UF-purified annatto and saffron for the preparation of UV protective cosmetic emulsions. Molecules. 23(9):2107.

Paula H. d, Pedrosa ML, Rossoni Júnior JV, Haraguchi FK, Santos R. C d, Silva ME. 2009. Effect of an aqueous extract of annatto (*Bixa orellana*) seeds on lipid profile and biochemical markers of renal and hepatic function in hipercholesterolemic rats. Braz Arch Biol Technol. 52(6): 1373–1378.

Pierpaoli E, Viola V, Barucca A, Orlando F, Galli F, Provinciali M. 2013. Effect of annatto-tocotrienols supplementation on the development of mammary tumors in HER-2/neu transgenic mice. Carcinogenesis. 34(6):1352–1360.

Raddatz-Mota D, Pérez-Flores LJ, Carrari F, Mendoza-Espinoza JA, de León-Sánchez FD, Pinzón-López LL, Godoy-Hernández G, Rivera-Cabrera F. 2017. Achiote (*Bixa orellana* L.): a natural source of pigment and vitamin E. J Food Sci Technol. 54(6):1729–1741.

Rattanavipanon W, Nithiphongwarakul C, Sirisuwansith P, Chaiyasothi T, Thakkinstian A, Nathisuwan S, Pathomwichaiwat T. 2021. Effect of tomato, lycopene and related products on blood pressure: A systematic review and network meta-analysis. Phytomedicine. 88:153512.

Reith J, Gielen J. 1971. Properties of bixin and norbixin and the composition of Annatto extracts. J Food Science. 36(6):861–864.

Rivera-Madrid R, Aguilar-Espinosa M, Cárdenas-Conejo Y, Garza-Caligaris LE. 2016. Carotenoid derivates in achiote (*Bixa orellana*) seeds: synthesis and health promoting properties. Front Plant Sci. 7:1406.

Roehrs M, Conte L, da Silva DT, Duarte T, Maurer LH, de Carvalho JAM, Moresco RN, Somacal S, Emanuelli T. 2017. Annatto carotenoids attenuate oxidative stress and inflammatory response after high-calorie meal in healthy subjects. Food Res Int. 100(Pt 1):771–779.

Santos G, Almeida M, Antunes L, Bianchi M. 2016. Effect of bixin on DNA damage and cell death induced by doxorubicin in HL60 cell line. Hum Exp Toxicol. 35(12):1319–1327..

Scotter M. 2009. The chemistry and analysis of annatto food colouring: a review. Food Addit Contam Part A. 26(8):1123–1145..

Shin J, Song M-H, Oh J-W, Keum Y-S, Saini RK. 2020. Pro-oxidant actions of carotenoids in triggering apoptosis of cancer cells: A review of emerging evidence. Antioxidants. 9(6):532–517.

Song B, Liu K, Gao Y, Zhao L, Fang H, Li Y, Pei L, Xu Y. 2017. Lycopene and risk of cardiovascular diseases: A meta-analysis of observational studies. Mol Nutr Food Res. 61(9):1601009.

Souza LF, Pagno CH, Medeiros N. d S, Barbosa S, dos Santos PCP, Rios A, Achaval M, de Jong EV. 2016. The effect of the carotenoid bixin and annatto seeds on hematological markers and nephrotoxicity in rats subjected to chronic treatment with cisplatin. Rev Bras Farmacogn. 26(4):446–450.

Sowmya PR-R, Arathi BP, Vijay K, Baskaran V, Lakshminarayana R. 2017. Astaxanthin from shrimp efficiently modulates oxidative stress and allied cell death progression in MCF-7 cells treated synergistically with β-carotene and lutein from greens. Food Chem Toxicol. 106(Pt A):58–69.

Suparmi Limantara L, Prasetryo B. 2008. Natural food colourant from seed of kesumba (*Bixa orellana* L.). In: International Conference on Food Science and Technology the Challenge of Universal Food Quality and Safety Regime. pp p. 1–10.

Sznarkowska A, Kostecka A, Meller K, Bielawski KP. 2017. Inhibition of cancer antioxidant defense by natural compounds. Oncotarget. 8 (9):15996–16016.

Tibodeau JD, Isham CR, Bible KC. 2010. Annatto constituent cis-bixin has selective antimyeloma effects mediated by oxidative stress and associated with inhibition of thioredoxin and thioredoxin reductase. Antioxid Redox Signal. 13(7):987–997.

Tinoush B, Shirdel I, Wink M. 2020. Phytochemicals: potential lead molecules for MDR reversal. Front Pharmacol. 11:832.

Ventosa E. 2018. *Bixa orellana* (annatto). In: CABI.org. https://www.cabi.org/isc/datasheet/9242. Accessed August 22, 2021.

Wang Y, Cui R, Xiao Y, Fang J, Xu Q. 2015. Effect of carotene and lycopene on the risk of prostate cancer: A systematic review and dose-response meta-analysis of observational studies. PLoS One. 10(9):e0137427.

Xiao H, Zheng Y, Ma L, Tian L, Sun Q. 2021. Clinically-relevant ABC transporter for anti-cancer drug resistance. Front Pharmacol. 12:648407.

Xu L, Li H, Wang Y, Dong F, Wang H, Zhang S. 2014. Enhanced activity of doxorubicin in drug resistant A549 tumor cells by encapsulation of P-glycoprotein inhibitor in PLGA-based nanovectors . Oncol Lett. 7(2):387–392.

Zechmeister L. 1960. Cis-trans isomeric carotenoid pigments. Fort Chem Org Naturst. 18: 224–349.

Bixin

ORIGINALITY REPORT

19%	16%	11%	4%
SIMILARITY INDEX	INTERNET SOURCES	PUBLICATIONS	STUDENT PAPERS

PRIMARY SOURCES

1	Submitted to Purdue University Student Paper	2%
2	www.mdpi.com Internet Source	2%
3	Vinícius J. Bolognesi, Carlos E.R. Garcia. "Annatto Carotenoids as Additives Replacers in Meat Products", Elsevier BV, 2018 Publication	2%
4	eprints.cmfri.org.in Internet Source	1%
5	pdfcoffee.com Internet Source	1%
6	jgpt.co.in Internet Source	1%
7	Rosanna Tarkany Basting, Pedro Manoel Barreto de Abreu, Ilza Maria de Oliveira Sousa, João Ernesto de Carvalho et al. " L. by-products' fractions from an industrial process: antiproliferative activity on tumor cells and chemical profile ", Natural Product Research, 2020	1%

	Publication	
8	oatao.univ-toulouse.fr Internet Source	1 %
9	coek.info Internet Source	1 %
10	ebin.pub Internet Source	1 %
11	mssrfcabc.res.in Internet Source	1 %
12	eprints.mercubuana-yogya.ac.id Internet Source	1 %
13	www.phcogrev.com Internet Source	1 %
14	Sukhchain Kaur, Harkomal Verma, Sharanjot Kaur, Subham Singh, Anil K. Mantha, Monisha Dhiman. "Chapter 270-1 Herbal Remedies for Improving Cancer Treatment Through Modulation of Redox Balance", Springer Science and Business Media LLC, 2022 Publication	1 %
15	Huan Xiao, Yongcheng Zheng, Lingling Ma, Lili Tian, Qiu Sun. "Clinically-Relevant ABC Transporter for Anti-Cancer Drug Resistance", Frontiers in Pharmacology, 2021 Publication	1 %
	aalborguh.rn.dk	

16	Internet Source	1 %
17	wiki2.org Internet Source	1 %
18	Beatriz de la Fuente, Gabriel López-García, Vicent Máñez, Amparo Alegría, Reyes Barberá, Antonio Cilla. "Antiproliferative Effect of Bioaccessible Fractions of Four Brassicaceae Microgreens on Human Colon Cancer Cells Linked to Their Phytochemical Composition", Antioxidants, 2020 Publication	1 %

Exclude quotes	On	Exclude matches	< 1%
Exclude bibliography	On		