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The Protective Effect of Dietary *Arthrospira* (*Spirulina*) *maxima* Against Mutagenicity Induced by Benzo[alpha]pyrene in Mice

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ABSTRACT Benzo[alpha]pyrene (B[α]P) was used to test the possible antimutagenic effects of *Arthrospira* (*Spirulina*) *maxima* (SP) on male and female mice. SP was orally administered at 0, 200, 400, or 800 mg/kg of body weight to animals of both sexes for 2 weeks before starting the B[α]P (intraperitoneal injection) at 125 mg/kg of body weight for 5 consecutive days. For the male dominant lethal test, each male was caged with two untreated females per week for 3 weeks. For the female dominant lethal test, each female was caged for 1 week with one untreated male. All the females were evaluated 13–15 days after mating for incidence of pregnancy, total *corpora lutea*, total implants and pre- and postimplant losses. SP protected from B[α]P-induced pre- and postimplant losses in the male dominant lethal test, and from B[α]P-induced postimplantation losses in treated females. Moreover, SP treatment significantly reduced the detrimental effect of B[α]P on the quality of mouse semen. Our results illustrate the protective effects of SP in relation to B[α]P-induced genetic damage to germ cells. We conclude that SP, owing mainly to the presence of phycocyanin, could be of potential clinical interest in cancer treatment or prevention of relapse.

KEY WORDS: • ***anticancer*** • ***antigenotoxic*** • ***antioxidant*** • ***blue-green algae*** • ***cyanobacterium***

INTRODUCTION

Arthrospira (*Spirulina*) *maxima* (SP) is a general term for plants that belong to Cyanophyceae.[1] It has been used as a food and nutritional supplement for a long time[2] and represents one of the richest sources of plant protein (60–70%) and a good source of vitamins, carotenoids, essential amino acids, minerals, and essential fatty acids (*e.g.,* γ-linolenic acid and sulfolipids).[3,4] Moreover, SP has phycocyanin, a photosynthetic biliprotein, and its chief phytochemical phycocyanobilin.[5]

In addition to the aforementioned nutrimental properties, SP is considered a nutraceutic based on the different pharmacological properties that have been reviewed by Belay,[6] Karkos *et al.*,[7] and Khan *et al.*[8] Recently, evidence was published about the effectiveness of SP in diabetic nephropathy[9] and liver injuries.[10] SP has also been shown to be effective against toxicity produced by cadmium,[11] arsenic,[12] mercuric chloride,[13] zinc,[11] and copper.[14] Moreover, SP has been effective against teratogenicity induced by hydroxyurea,[15] cancer in humans and animals,[16,17] and mutagenicity.[18] These properties could be due, in part, to the antioxidant capacity of the algae as a whole,[10,19] its protean and water extracts,[20,21] or bioactive components (such as phycocyanin).[22] Moreover, it was demonstrated that SP did not produce genotoxicity in CD1 mice, using the dominant lethal test and sperm abnormalities assay.[23]

Benzo[alpha]pyrene (B[α]P), a prototypical mutagenic and carcinogenic polycyclic aromatic hydrocarbon, is a product of incomplete combustion or pyrolysis of organic material.[24] It is found ubiquitously in cigarette smoke, urban air, charbroiled food, vehicle exhaust, asphalt, and coke ovens.[25,26] It is a complete carcinogen, acting in both the initiation and growth promotion stages of chemical carcinogenesis,[27] and it is thought to have negative effects on male reproduction,[28] the immune system, embryonic development, and spermatogonial cells.[28–31]

Some studies have demonstrated that the genotoxicity of this polycyclic aromatic hydrocarbon is highly associated with oxidative stress and DNA damage by electrophilic attack, thus decreasing the activity of endogenous antioxidants, while increasing the level of lipid peroxidation, reactive oxygen species, glutathione, and the percentage of cells in the G0/G1 phase.[29,32] These effects are largely due

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to the metabolic activation of this hydrocarbon, principally triggered by cytochrome P450 (CYP450) enzymes (CYP1A1 and CYP1B1) and resulting in the formation of the reactive B[α]P-7,8-dihydrodiol-9,10-epoxide (BPDE). This in turn leads to DNA miscoding adduct formation and subsequent mutations, which may initiate carcinogenesis (B[α]P).[33,34]

B[α]P and its metabolites also modulate mammalian gene expression through epigenetic mechanisms involving the aryl hydrocarbon receptor signaling pathway and altered mitogenic signaling, to contribute to the tumor-promoting effects.[29,35] Moreover, changes in microRNA-mRNA expression in human cells, part of the regulation of mammalian cell functions, have been used to identify molecular mechanisms of genotoxicity of B[α]P.[36,37]

For the last few years, there has been an increasing interest in carrying out *in vivo* and *in vitro* studies on the use of antigenotoxic agents for cancer treatment. Such agents include a wide variety of diets, plants, mushrooms, and algal extracts.[38–42] These agents can block, inhibit, reverse, or retard the process of carcinogenesis through various processes, such as antioxidative, anti-inflammatory, induction of phase II enzymes, apoptosis, and cell cycle arrest.[43]

The aim of the present study was to evaluate the effects of SP on B[α]P, an indirect mutagen, in germ cells of male and female mice, using the dominant lethal test and the sperm abnormalities assay.

MATERIALS AND METHODS

Animal husbandry and maintenance

Male and virgin female CF1 mice, 9–10 weeks old, were obtained from the Virology Institute Animal House (Secretary of Health, Mexico City).

The animals were housed in polycarbonate cages in an air-conditioned room (22°C ± 1°C, 50–60% relative humidity) with a photoperiod of 12 h, from 8 am to 8 pm. They were fed Purina rodent laboratory chow and tap water *ad libitum*. All the animals were acclimatized for at least 7 days before use. They were handled according to the Institutional Guidelines and Mexican Official Standard (NOM-062-ZOO-1999) regarding technical specifications for production, care, and use of laboratory animals. All the studies were approved by the Bioethical Committee of the National School of Biological Sciences (approval document ENCB/CBE/101/10/07) on October 3, 2007.

Test substances

B[α]P used in this study was purchased from Sigma-Aldrich Chemical Co. (St. Louis, MO, USA) and stored at room temperature. SP powder was from a bulk production batch supplied by Alimentos Esenciales Para la Humanidad, S.A. de C.V. (Mexico City, Mexico).

Experimental protocol

This study was conducted in two phases: a male dominant lethal phase and a female dominant lethal phase.[44,45] In both cases, male and female animals were randomly divided into seven groups: (I) Control oil, (II) Control water, (III) B[α]P 125 mg/kg, (IV) SP 800 mg/kg, (V) B[α]P 125 mg/kg + SP 200 mg/kg, (VI) B[α]P 125 mg/kg + SP 400 mg/kg, and (VII) B[α]P 125 mg/kg + SP 800 mg/kg.

SP was administered through gastric intubation at doses of 0 (water), 200, 400, or 800 mg/kg, under a consistent volume factor of 10 mL/kg of body weight for 2 weeks before starting the B[α]P treatment. The dosage range was consistent with the doses employed in other studies on the antigenotoxicity of SP in mice.[46,47]

B[α]P was suspended in corn oil at a concentration of 12.5 mg/mL immediately before use and intraperitoneally injected at a daily dose of 125 mg/kg of body weight for 5 days. This dose is known to induce dominant lethal mutations in mice.[48] We employed the most widely used approach for male dominant lethal studies to evaluate mutational effects at particular developmental stages of male germ cells,[49–51] which is a daily injection for 5 consecutive days.

Male dominant lethal test

In this test, all 13 males, with or without SP treatment, were housed with two naive, nulliparous females overnight for 7 days (days 1–7) and then were again housed overnight with different females for 7 days (days 7 to 14 and days 15 to 21). Mating was determined by the presence of a vaginal plug on the morning after cohabitation. The day on which mating was detected was designated as gestation day 0, at which time the pregnant female was transferred to another cage. Cohabitation continued until copulation was detected.

Female dominant lethal test

In the female dominant lethal test, 10 untreated males were mated with 20 treated females (1–2) in each cage for 1 week, as previously described.

In both the male and female dominant lethal test, mated females were observed daily for signs of toxic effects. They were sacrificed on gestational day 13–15 by CO_2 asphyxiation and cervical dislocation. A laparatomy was performed to observe the uterus and ovaries. The number of pregnant females, *corpora lutea*, implantation sites, and dead implants were recorded. The frequency of pre- and postimplantation losses was estimated. These data were analyzed for statistical significance against their respective controls by using the chi-square test, with significance set at $P < .05$. The protective index was calculated.

Sperm abnormalities

In each of the seven groups of male treated mice, six animals were sacrificed 1, 3, and 5 weeks after the last injection. The distal portion of the right epididymis and the *vas deferens* were immediately dissected by cutting the middle tail section and immersing it in 1 mL of saline solution at 37°C. A syringe with an additional 1 mL of the same solution was inserted into the previously opened *vas deferens* to perfuse the tissues. Then, the semen suspension was stirred and thoroughly mixed by repeated pipetting. The sperm concentration, motility, and morphology of each

animal were assessed after homogenization as described by Albert and Roussel.[52] Counts were made with a Thomas hemocytometer and results were expressed in millions of spermatozoa/mL of suspension. One drop of smear was mounted between a slide and a coverslip and was immediately examined to determine motility. The results are expressed in percentages. One hundred spermatozoa from each animal were analyzed and classified as either normal or abnormal. Head, midpiece, and main piece abnormalities were recorded. Finally, the left testis, epididymis, and seminal vesicles were weighed in the group sacrificed 5 weeks after the end of the B[α]P treatment. Testes histological analysis was not done. Data on sperm morphology, motility, and epididymal sperm count were analyzed by using the Mann–Whitney *U*-test. Data on sex organ weights were analyzed by the Student's *t*-test. Results were considered significant at $P < .05$. All analyses were performed by using the statistical software SigmaStat® version 2.03.

RESULTS

Male dominant lethal test

Observations from this test are summarized in Table 1. Mating and pregnancy rates of females mated with B[α]P-treated males were high during 3 weeks of cohousing, with no difference found in relation to control values. However, preimplantation and postimplantation losses were significantly higher ($P < .05$) in the females mated with males treated only with B[α]P, in each of the three periods of cohabitation: 18.2, 16.8, and 19.8 for preimplantation and 19.5, 17.2, and 18.9 for postimplantation losses during the first, second, and third week, respectively.

However, in females mated with B[α]P + SP-treated males, preimplantation and postimplantation losses during the first, second, and third week of mating were found in the same range as in the control group. Consequently, the protective index during each week was 35.6, 33.1, and 39.4 at the 200 mg/kg dose, 47.8, 24.9, and 53.5 at the 400 mg/kg dose, and 77.3, 69.7, and 65.6 at the 800 mg/kg dose, respectively. SP protected against the B[α]P-induced genotoxicity in a dose-dependent manner. There was no significant change in any of the parameters of genotoxicity in animals treated with SP alone at the 800 mg/kg dose.

Female dominant lethal test

Table 2 shows that mating and pregnancy rates of B[α]P-treated females were also not affected. Whereas preimplantation losses in B[α]P-treated females were similar to those in the control, the frequency of

TABLE 1. REPRODUCTIVE DATA FOR UNTREATED FEMALE MICE MATED TO MALES TREATED WITH *SPIRULINA* AND BENZO[ALPHA]PYRENE

Dose (mg/kg)	*Mating interval (days)*	*No. of mated females*	*No. of pregnant females (%)*	*Total corpora lutea*	*Total implants*	*Frequency of preimplant loss*[a]	*Dead implants*	*Frequency of postimplant loss*[b]	*Protective index (%)*[c]
Control oil	1–7	26	22 (84.6)	229	200	12.6	19	9.5	—
	8–14	26	24 (92.3)	290	254	12.4	25	9.8	—
	15–21	26	23 (88.5)	266	235	11.6	29	12.3	—
Control water	1–7	26	23 (88.4)	282	250	11.1	29	11.8	—
	8–14	26	24 (92.3)	280	245	12.2	26	10.4	—
	15–21	26	24 (92.3)	302	264	12.8	38	14.5	—
B[α]P	1–7	26	20 (79.6)	250	204	18.2*	40	19.5*	—
	8–14	26	21 (80.8)	231	192	16.8**	48	17.2***	—
	15–21	26	21 (80.8)	237	190	19.8***	36	18.9*	—
SP800	1–7	26	26 (100)	300	271	9.7	28	10.2	—
	8–14	26	23 (88.4)	271	241	11.1	24	9.8	—
	15–21	26	23 (88.4)	264	230	12.9	26	11.3	—
B[α]P + SP200	1–7	26	24 (92.3)	281	249	11.4†	31	12.6	35.6
	8–14	26	25 (96.1)	310	278	10.3†	32	11.5†††	33.1
	15–21	26	22 (84.6)	264	238	9.8	24	10.2†	39.4
B[α]P + SP400	1–7	26	24 (92.3)	283	253	10.5†	30	11.7†	47.8
	8–14	26	21 (80.8)	233	210	9.9†	22	10.4†††	24.9
	15–21	26	23 (88.5)	278	251	9.6	25	10.0††	53.5
B[α]P + SP800	1–7	26	24 (92.3)	273	243	11.1†	23	9.6††	77.3
	8–14	26	25 (96.1)	243	217	10.7	20	9.2†††	69.7
	15–21	26	22 (84.6)	305	274	10.1	22	8.1†††	65.6

[a]Frequency of preimplant loss = [(total corpora lutea − total implants)/total corpora lutea] × 100.
[b]Frequency of postimplant loss = (dead implants/total implants) × 100.
[c]Protective index (%) = 100 − [(frequency of postimplant loss SP + B[α]P treated)/frequency of postimplant loss B[α]P treated] × 100.
Calculations were based on absolute figures and compared at the corresponding weeks. Those marked with asterisks differ significantly from the control oil value: *$P < .05$, **$P < .01$, ***$P < .001$. Those marked with daggers differ significantly from the B[α]P value: †$P < .05$, ††$P < .01$, †††$P < .001$.
B[α]P, benzo[alpha]pyrene (125 mg/kg, intraperitoneally); SP, *Spirulina* (200, 400, or 800 mg/kg by gastric intubation).

Table 2. Reproductive Data for Female Mice Treated with *Spirulina* and Benzo[alpha]pyrene Mated to Untreated Mice

Dose (mg/kg)	*No. of mated females*	*No. of pregnant females (%)*	*Total corpora lutea*	*Total implants*	*Frequency of preimplant losses*[a]	*Dead implants*	*Frequency of postimplant losses*[b]	*Protective index (%)*[c]
Control oil	20	17 (85)	195	170	12.8	15	8.8	—
Control water	20	18 (90)	225	198	12.2	16	8.0	—
B[α]P	20	15 (75)	165	135	13.2	23	16.5†††	—
SP800	20	18 (90)	218	195	10.4	17	8.8	—
B[α]P + SP200	20	17 (85)	187	166	11.3	14	9.2†	44.3
B[α]P + SP400	20	16 (80)	160	150	12.5	12	8.7†	47.3
B[α]P + SP800	20	16 (80)	184	162	11.8	11	7.2††	56.4

[a]Frequency of preimplant loss = [(total corpora lutea − total implants)/total corpora lutea] × 100.
[b]Frequency of postimplant loss = (dead implants/total implants) × 100.
[c]Protective index (%) = 100 − [(frequency of postimplant loss SP + B[α]P treated)/frequency of postimplant loss B[α]P treated] × 100.
Calculations were based on absolute figures. Those marked with daggers differ significantly from the B[α]P value: †$P < .05$; ††$P < .01$; †††$P < .001$.

postimplantation losses was significantly different ($P < .05$), reaching 16.5% in the experimental group. However, pretreatment with 200, 400, and 800 mg/kg doses of SP significantly reduced the frequency of postimplantation losses to 9.2, 8.7, and 7.2, respectively, representing a protective index of 44.3, 47.3, and 56.4, respectively. As in the male dominant lethal test, SP alone did not induce a mutagenic effect *per se*.

Table 3. Epididymal Sperm Concentration, Motility, and Percentage of Morphologically Abnormal Sperm in Mice Treated with *Spirulina* and Benzo[alpha]pyrene

Dose (mg/kg)	*Week 1*	*Week 3*	*Week 5*
Sperm concentration ($\times 10^6$ mL)			
Control oil	7.3 ± 1.5	7.0 ± 1.1	8.2 ± 1.6
Control water	7.9 ± 1.2	7.6 ± 0.9	8.0 ± 1.3
B[α]P	7.0 ± 1.0	5.8 ± 1.2*	5.7 ± 1.2*
SP800	7.6 ± 1.2	7.9 ± 1.2	7.6 ± 1.5
B[α]P + SP200	6.5 ± 1.1	6.1 ± 1.4	6.3 ± 1.0
B[α]P + SP400	6.9 ± 1.4	7.7 ± 1.6†	7.9 ± 1.4†
B[α]P + SP800	7.1 ± 1.3	7.5 ± 1.3†	8.1 ± 1.6††
Reduced sperm motility (%)			
Control oil	15.9 ± 3.5	16.0 ± 2.5	15.8 ± 3.4
Control water	16.7 ± 3.8	15.4 ± 3.3	16.3 ± 3.7
B[α]P	15.8 ± 4.2	14.8 ± 2.9	19.8 ± 3.2*
SP800	16.2 ± 4.4	15.1 ± 2.7	16.2 ± 4.3†
B[α]P + SP200	16.5 ± 4.6	15.2 ± 3.0	15.7 ± 3.9†
B[α]P + SP400	15.7 ± 3.6	16.1 ± 3.6†	16.0 ± 3.8†
B[α]P + SP800	16.0 ± 4.5	15.6 ± 3.1	16.4 ± 4.1†
Normal shaped sperm (%)			
Control oil	71.1 ± 4.2	68.9 ± 4.0	66.6 ± 4.8
Control water	70.2 ± 5.1	69.6 ± 3.9	65.6 ± 4.7
B[α]P	71.6 ± 5.0	69.2 ± 4.1	65.7 ± 4.2
SP800	68.4 ± 4.8	70.5 ± 4.6	68.2 ± 5.0
B[α]P + SP200	68.8 ± 4.7	68.7 ± 4.0	60.2 ± 3.8
B[α]P + SP400	70.1 ± 5.3	70.3 ± 3.7	64.8 ± 3.5
B[α]P + SP800	69.9 ± 5.4	68.8 ± 3.7	63.9 ± 3.9

Values are mean ± stardard error.
Those marked with asterisks differ significantly from the control oil value: *$P < .05$. Those marked with daggers differ significantly from the B[α]P value: †$P < .05$; ††$P < .01$.

Sperm abnormalities

The toxic effect of B[α]P and protective effect of SP on epididymal sperm is shown in Table 3. B[α]P treatment of male mice caused a significant decrease ($P < .05$) in sperm concentrations at 3 and 5 weeks, as well as in motility at 5 weeks after treatment. Treatment at the two higher doses of SP significantly reduced these effects. No changes were observed in the normal shape of the sperm.

The testis, epididymis, and seminal vesicle weights significantly decreased ($P < .05$) in the group treated only with B[α]P (Table 4). However, the treatment with 400 and 800 mg/kg of SP significantly ($P < .05$) attenuated the weight loss of the testis and epididymis. The weight of seminal vesicles was also significantly improved with SP treatment.

DISCUSSION

Dominant lethal mutations are genetic changes induced in parent germ cells that cause the death of the resulting zygote, its failure to implant, or early embryonic death if implanted.[53] The dominant lethal test is the primary germ cell mutagenicity assay for determining the existence of dominant lethal mutations.[54] Studying the increase of sperm abnormalities in mice is also a good model for detecting possible genotoxicity caused by chemical agents.[55]

The mating rate observed in the current study, whether with treated males or treated females, was not significantly different compared with that observed in the control group, indicating that there was no decrease in the pregnancy of the animals in the experimental groups. In untreated female mice mated with B[α]P-treated males, pre- and postimplantation losses were significantly increased in each of the three periods of cohabitation. On the other hand, in B[α]P-treated females mated with untreated males, only postimplantation losses were significantly increased. These results suggest that in B[α]P-treated females, lesions were induced in the mature dictyate oocyte[56] and that both sexes were similarly sensitive to B[α]P toxicity.

Table 4. Final Body and Sex Organ Weight of Male Mice Sacrificed After the Completion of treatment with Benzo[α]pyrene (i.p.) + *Spirulina* (I.G.)

Dose (mg/kg)	*Terminal body weight (g)*	*Testis (g)*	*Epididymis (mg)*	*Seminal vesicles (mg)*
Control oil	35.8 ± 3.7	0.598 ± 0.06	58.1 ± 2.8	0.20 ± 0.02
Control water	36.6 ± 4.0	0.601 ± 0.07	58.7 ± 4.0	0.18 ± 0.03
B[α]P	35.6 ± 3.8	0.507 ± 0.08*	48.8 ± 3.0*	0.12 ± 0.01*
SP800	36.0 ± 4.2	0.611 ± 0.05	57.8 ± 4.1	0.15 ± 0.04
B[α]P + SP200	36.2 ± 4.5	0.550 ± 0.04	50.5 ± 3.6	0.22 ± 0.04††
B[α]P + SP400	34.9 ± 3.9	0.604 ± 0.05†	60.4 ± 4.4†	0.19 ± 0.03†
B[α]P + SP800	37.0 ± 3.6	0.614 ± 0.07†	58.7 ± 3.9†	0.19 ± 0.03†

Values are mean ± standard error.

Those marked with asterisks differ significantly from the control oil value: *$P < .05$. Those marked with daggers differ significantly from the B[α]P value: †$P < .05$; ††$P < .01$.

The three post-treatment periods after B[α]P application account for the postmeiotic stages of spermatogenesis, where germ cell stages in males were found to be most sensitive to damaged induction.[57] Days 1–7 correspond to epididymal spermatozoa, days 8–14 to testicular spermatids, and days 15–21 to early spermatids.[58]

Treatment with SP alone at 800 mg/kg did not induce dominant lethality in either males or females. This result is also in agreement with previous reports, where rats and mice were given the algae at various concentrations in their diet to test for genotoxicity.[23,59]

In the different groups treated with SP + B[α]P, the prevention of genotoxicity was evidenced by a significant decrease in the frequency of pre- and postimplantation losses (with the exception of the first week of the animals that received 200 mg/kg of SP, Table 1), as compared with the control group (B[α]P) only treatment, and therefore by an increase in the protective index, calculated based on the frequency of postimplantation losses.

The protective index of SP was generally dose dependent and was above that reported for natural products such as black tea (also employing the male and female dominant lethal test).[60] The greatest protective effect (77%) was found with the dose of 800 mg/kg of SP during the mating interval period, which corresponds to epididymal spermatozoa.

Antimutagenic protection of SP has also been found against cyclophosphamide, employing the rodent dominant assay in mice[47] and against mitomycin-C, cisplatin and urethane, marrow, using the micronucleous test.[18,46] Moreover, a radioprotective effect in mice has been reported.[61] Additionally, the *Tradescantia* bioassay provides evidence of the anticlastogenic properties of this cyanobacterium.[62] Other natural products and phytochemicals have shown comparative mutagenic protection against B[α]P, based on their antidoxidant activity, capture of free radicals, modulation of cell metabolism, stimulation of apoptosis, or other mechanisms.[60,63–67]

The antimutagenic and anticarcinogenic effects of SP have been attributed, in many *in vitro* and *in vivo* studies, to its antioxidant and antiapoptotic capacity.[68] Indeed, the capacity of SP to reduce levels of lipid peroxidation has been demonstrated, levels that are increased by B[α]P treatment.[69] There are also reports on its capacity to increase the enzymatic activity of antioxidant biomarkers (superoxide dismutase, catalase, glutathione peroxidase, glutathione-transferase),[13,70] activity that is decreased by B[α]P treatment.[69]

The antioxidant properties can be attributed to bioactive compounds found in SP, such as carotenoids (especially of β-carotene), omega 3, polyunsaturated fatty acids, provitamins, and other vitamins (including A, B, and E).[21,71,72] However, a great majority of studies demonstrate that the principal molecule responsible for this property is phycocyanin. This substance has shown a powerful antioxidant and antiproliferative activity against human cancer cells through apoptosis and nuclear apoptosis induction accompanied by G0/G1 phase arrest and DNA fragmentation.[73] Phycocyanin alone has significant free radical scavenging[74–77]and anticancer effects[78,79] and can be combined with other anticancer drugs, such as topotecan.[80] On the other hand, in combination with piroxicam, phycocyanin can stimulate cytochrome C release by downregulating the Bcl-2 (an antiapoptotic protein) expression, thus mediating mitochondrial-dependent apoptosis in DMH-induced colon cancer.[81]

Previous reports of decreased epididymal sperm concentration and motility after treatment of rats with B[α]P[47] were confirmed by the current study. This could be due to a direct effect on the spermatogenic compartment. Our study demonstrated that SP treatment substantially buffered B[α]P-induced testicular spermatogenic cell damage, providing further evidence of its antigenotoxic activity.

SP belongs to one of the few algal species that has a proven antioxidant activity and scavenging effect.[19] It is probable that phycocyanin and various components are involved as studies have shown that the synergistic action of a wide spectrum of antioxidants is more efficient than only one such agent. Furthermore, it is known that antioxidants from natural sources (primarily food) have a higher bioavailability and therefore a higher protective efficacy than synthetic ones.[82]

We conclude that SP exhibited a protective effect against B[α]P-induced genetic damage to germ cells in male and female mice, and may serve as a natural agent in cancer treatment or chemoprevention.

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AUTHOR DISCLOSURE STATEMENT

No competing financial interests exist.

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