Original Article

Mini Series on microbial and dental problems

Studies on Phytoconstituents, *In vitro* Antioxidant, Antibacterial, and Cytotoxicity Potential of *Argemone mexicana Linn*. (Family: *Papaveraceae*)

Kailas D. Datkhile, Satish R. Patil, Madhavi N. Patil, Pratik P. Durgawale, Nilam J. Jagdale, Vinit N. Deshmukh

Department of Molecular Biology and Genetics, Krishna Institute of Medical Sciences "Deemed to be University", Satara, Maharashtra, India

Abstract

Background: The present study was designed to determine the antioxidant, antimicrobial, and cytotoxic activity of *Argemone mexicana*. **Materials and Methods:** Aqueous, methanol, and ethanol extracts of the whole plant *A. mexicana* were screened for phenolics, tannins, and flavonoids. The *in vitro* antioxidant activity was evaluated through 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid), 2,2-diphenyl-1-picryl -hydrazyl-hydrate, and ferric reduction activity potential assays. The antimicrobial activity of the extracts against *Klebsiella pneumoniae*, *Escherichia coli, Pseudomonas aeruginosa*, and *Staphylococcus aureus* was determined. Cytotoxic properties of the extracts were studied using the cancer cell lines HeLa, MCF-7, and HCT-15 through cell viability and DNA fragmentation assays. **Results:** Phytochemical analysis revealed that the extracts contained phenolics, tannins, and flavonoids. The aqueous and solvent extracts of the whole plant exhibited a strong antioxidant activity *in vitro* and antibacterial activity against the tested human pathogenic bacteria. The crude extract of *A. mexicana* exhibited a strong cytotoxic activity against the tested cancer cell lines. **Conclusion:** Phytoconstituents from the crude extract of *A. mexicana* exhibited higher antioxidant, antibacterial, and cytotoxic activities than earlier reported annotations.

Keywords: Antioxidants, *Argemone mexicana*, cytotoxicity, genotoxicity, phytoconstituents

Introduction

Medicinal plants are enriched with a wide range of natural phytochemicals, and these phytochemicals have been extensively studied in recent years for their imperative remedial relevance in the therapeutics of deadly ailments. The World Health Organization has quoted that approximately 80% of the global population conventionally relies on natural flora for their healthcare needs. Conentionally, the rural and semi-urban parts of Indian, as well as other subcontinents, utilize diverse flora in their medical remedies. Phytoconstituents of several floras have attained certain level of importance in both homeopathy and Ayurveda owing to their various traditional applications, but the pharmaceutical value of many plant species remain unexplored. Therefore, researchers are now interested in identifying and elucidating the mechanisms of action of phytoconstituents of several medicinal plants in the treatment of different medical conditions. Numerous studies have revealed the antioxidant,[1,2] antimicrobial,[3,4] and anticancer[5,6] properties of medicinal plants, but the biological potential of several plants remain unexplored.

Argemone mexicana, commonly known as the Mexican poppy plant (Family: Papaveraceae), was reported to have multiple remedial benefits in traditional medicine for the treatment of numerous conditions, including cough, asthma, jaundice, ulcers, warts, skin diseases, and leprosy.[7,8] *A. mexicana* is a toxic weed widely distributed throughout the arid belts of Maharashtra, Karnataka, and the Western Ghats in India. Other studies have investigated the abundant phytoconstituents such

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Address for correspondence: Dr. Kailas D. Datkhile, Department of Molecular Biology and Genetics, Krishna Institute of Medical Sciences (Deemed to be University), Karad, Satara - 415 110, Maharashtra, India.
E-mail: hodgeneticslab@kimsuniversity.in

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as alkaloids and phenolics concealed in different plant parts of the Papaveraceae members.[9,10] Studies have also reported the biological potential of phytoconstituents from different parts of *A. mexicana*, including free radical scavenging,[11] antibacterial,[12] and antiparasitic activities.[13] Attempts have been made to demonstrate the antiproliferative activity of phytoconstituents from different parts of *A. mexicana.* However, higher concentrations of plant extracts inhibit cell growth to a minimum extent in different cancer cell lines.[14,15] Earlier studies on the restorative properties of plant chemicals of *A. mexicana* have diversified and contradictory opinions. Therefore, in the present study, *A. mexicana* was used to investigate the effect of the different solvent extractions on phytochemicals and to evaluate the antioxidant and antibacterial properties of different solvent extracts of the whole plant. This study was further extended to determine the cytotoxic and genotoxic potential of aqueous, methanol, and ethanol extracts of *A. mexicana*.

Materials and Methods

Collection and processing of plant materials

A. mexicana plants were collected from the arid region of the Western Ghats of southwestern Maharashtra, India. The whole plants were thoroughly washed with distilled water for removing adhered dust particles, dried, and then ground into a fine powder using mortar and pestle.

Preparation of solvent extracts

Exactly 25 gram (g) of plant powder was used for soxhlet extraction with solvents, namely distilled water, methanol, and ethanol. The methanol extraction displayed 20% yield, and ethanol extraction yielded 16%, whereas aqueous extract produced 40% yield after 15 cycles of extraction. Thereafter, the dried powder dissolved in respective solvents to get defined concentration (10 mg/mL) and used for further experiments.

Phytochemical screening of *Argemone mexicana* plant extracts

Total polyphenolic, tannin, and flavonoid contents in aqueous, methanol, and ethanol extracts of the whole plant were determined using the method described by Durgawale *et al.*;[16] gallic acid, tannic acid, and rutin were used as standards.

Evaluation of antioxidant properties of *Argemone mexicana* plant extracts

The antioxidant potential of aqueous, methanol and ethanol extracts of *A. mexicana* was determined through free radical scavenging assays such as 2,2′-azino -bis(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS), 2,2-diphenyl-1-picryl-hydrazyl-hydrate (DPPH), superoxide radical scavenging, and ferric reducing antioxidant power assays, as described earlier by Durgawale *et al.*[16]

Evaluation of antimicrobial properties of *Argemone mexicana* plant extracts

The antimicrobial activity of plant extracts was tested against the Gram-positive bacterium *Staphylococcus aureus* (ATCC® 29213™) and the Gram-negative bacteria *Escherichia coli* (ATCC® 25922™), *Klebsiella pneumoniae* (ATCC® 700603™), and *Pseudomonas aeruginosa* (ATCC® 2617™) by using the minimum inhibitory concentration method.

In vitro evaluation of cytotoxicity activity of *Argemone mexicana* plant extract

The cytotoxic effect of *A. mexicana* plant extracts on human cancer cell lines, namely HeLa, MCF-7, and HCT-15 cell lines, was evaluated through the 3-(4,5-dimethylthiazole -2-yl)-2,5-diphenyltetrazolium bromide (MTT) assay. In a 96-well plate, 10,000 cells were added to 100 µL of minimum essential medium and incubated at 37°C and 5% CO_2. After 24 h of incubation, confluent cells were exposed to the extracts at concentrations of 0.010, 0.025, 0.050, 0.1, and 0.2 mg/mL in a culture medium without fetal bovine serum and were further incubated for 48 h at 37°C and 5% CO_2. Cell viability was observed after every 24-h interval. After the completion of 24-h and 48-h treatments, the cells were treated with MTT (5 mg/mL, 10 µL/well). After 4 h of incubation, the MTT solution was removed, and 200 µL dimethyl sulfoxide was added to each well. The absorbance of the developed purple color was measured at 570 nm

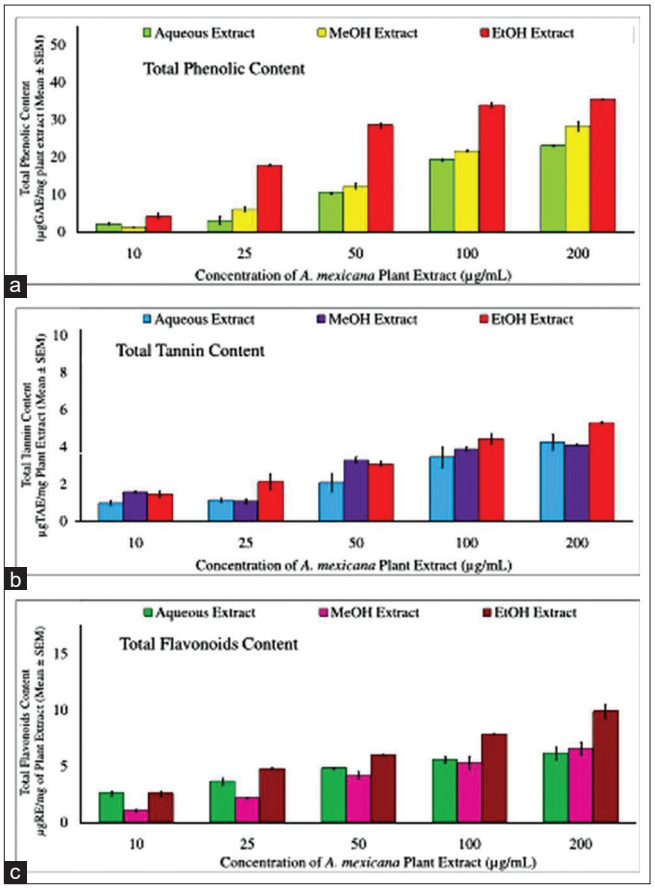

Figure 1: Representative histogram showing (a) total phenolic content, (b) total tannin content and (c) flavonoids content in the aqueous, methanol, and ethanol extracts of *Argemone mexicana* L. plant. Values are expressed as the means of three independent experiments

by using an ultraviolet-visible 1800 spectrophotometer (Shimadzu). The morphology of the cells was then examined using an inverted phase-contrast microscope (Primovert Carl Zeiss).

DNA fragmentation assay

The genotoxic activity of the *A. mexicana* extracts was determined through a DNA fragmentation assay, which was performed according to the procedure described by Durgawale *et al*.[16] In this assay, HeLa, MCF-7, and HCT -15 cells (1×10^6) were treated with 10, 25, 50, 100, and 200 µg/mL concentrations of aqueous, methanol, and ethanol extracts of the plant.

Statistical analysis

All experiments were performed in triplicates, and values were expressed as mean ± standard error of three measurements. Statistical analysis was carried out using Microsoft Excel 2010 to evaluate the possible differences among the means.

Results

Phytochemical screening of *Argemone mexicana* solvent extracts

The primary phytochemical screening of the whole plant *A. mexicana* using different solvents indicated the presence of various constituents, including phenolics, tannins, and flavonoids. The results were obtained through colorimetric quantitations by using the standard curves of gallic acid, tannic acid, and rutin as standards for phenolics, tannins, and flavonoids, respectively. The total phenolic, tannin, and flavonoid content of *A. mexicana* is outlined in Figure 1a-c, respectively.

In vitro radical scavenging activity of *Argemone mexicana* plant extracts

The *in vitro* radical scavenging activity of different *A. mexicana* solvent extracts is presented in Figure 2a-d. The highest ABTS radical scavenging activity was observed in 200 µg/mL of the ethanol extract, followed by the methanol and aqueous extracts (89.51 ± 0.81, 86.30 ± 1.18, and 77.71 ± 0.25, respectively) [Figure 2a]. The concentration required to inhibit 50% of the ABTS radical activity (IC_{50}) was 73.61 ± 1.87 µg/mL for the aqueous extract, 67.84 ± 1.43 µg/mL for the methanol extract, and 63.92 ± 2.02 µg/mL for the ethanol extract. Higher DPPH radical scavenging activity was observed in 200 µg/mL of the ethanol (93.12 ± 4.09) and methanol (84.51 ± 0.49) extracts than in the aqueous extract (77.71 ± 3.01) of *A. mexicana* [Figure 2b]. The IC_{50} value for the inhibition of DPPH radicals was 68.76 ± 2.87 µg/mL for the aqueous extract, 50.66 ± 3.48 µg/mL for the methanol extract, and 34.39 ± 3.32 µg/mL for the ethanol extract. The superoxide radical scavenging activity of the aqueous extract (81.87 ± 0.34) was lower than that of the methanol (82.90 ± 1.51) and ethanol (91.20 ± 0.34) extracts [Figure 2c]. The IC_{50} value for the inhibition of superoxide radicals was 50.13 ± 1.96 µg/mL for the aqueous extract, 52.61 ± 3.95 µg/mL for the methanol extract, and 61.90 ± 1.09 µg/mL for the ethanol extract.

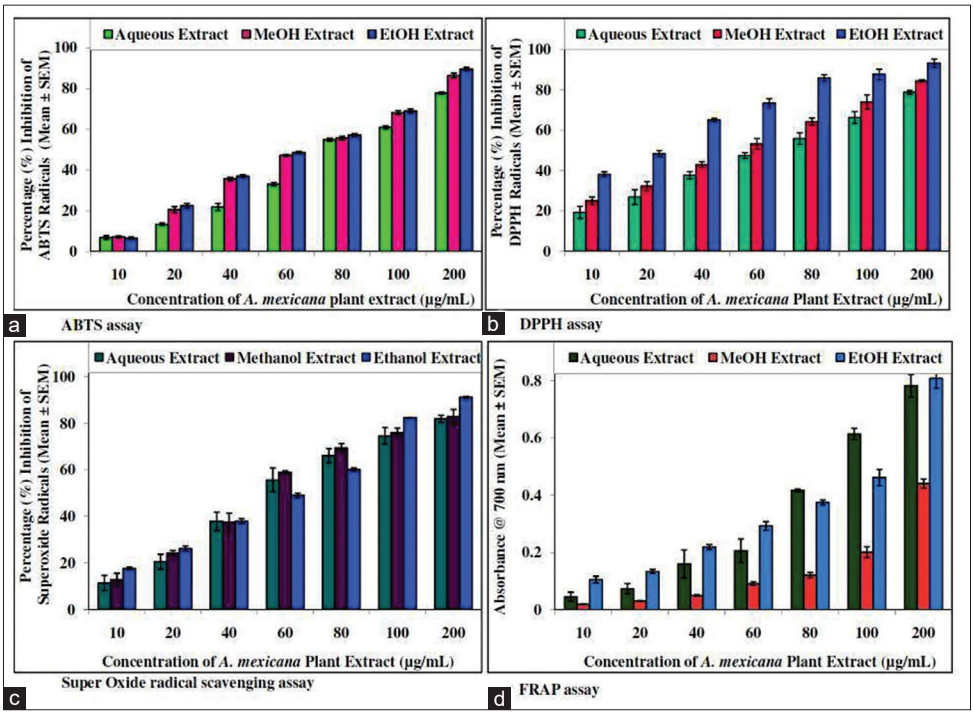

Figure 2: Representative histogram showing (a) 2,2′-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) and (b) 2,2-diphenyl-1-picryl-hydrazyl-hydrate (c) Superoxide radical scavenging activity and (d) ferric reduction activity potential assay of different concentrations of aqueous, methanol and ethanol extracts of *Argemone mexicana.* The data represent the percentage of inhibition of radicals *in vitro*. The results represent the means of three independent experiments, and error bars represent the standard error of the means

Figure 2d illustrates the reducing capacity of selected solvent extracts of *A. mexicana*, which increased gradually with an increase in the concentration of the extract.

Antimicrobial potential of *Argemone mexicana* plant extracts

The antibacterial potential of the aqueous, methanol, and ethanol extracts of *A. mexicana* against the pathogenic bacteria *S. aureus, K. pneumoniae, E. coli,* and *P. aeruginosa* was studied using the minimum inhibitory concentration method. Among the tested solvent extracts, the ethanol extract exhibited maximum antimicrobial activity against the selected bacterial species. The *A. mexicana* aqueous extract inhibited the growth of *E. coli* cells by 56.87% ± 1.75%, whereas the methanol and ethanol extracts inhibited the growth of *E. coli* cells by 58.22% ± 60.96% and 62.67% ± 0.27%, respectively, at a concentration of 2 mg/mL [Figure 3a]. Similarly, the aqueous, methanol, and ethanol extracts of *A. mexicana* inhibited the growth of *S. aureus* cells by 35% ± 1.29%, 66.10% ± 0.71%, and 77.54% ± 1.19%, respectively [Figure 3d]. Lower inhibition of cell growth was observed for *P. aerugionosa* (34.68% ± 1.51% by the aqueous extract, 36.16% ± 1.40% by the methanol extract, and 40.97% ± 1.42% by the ethanol extract) [Figure 3b] and *K. pneumonia* (30.41% ± 1.40% by the aqueous extract, 33.14% ± 1.06% by the methanol extract, and 38.28% ± 0.54% by the ethanol extract) [Figure 3c] when exposed to 2 mg/mL of the *A. mexicana* extract.

Cytotoxicity and genotoxicity activity of *Argemone mexicana* plant extract

The percentage viability of HeLa cells exposed to the aqueous, methanol, and ethanol extracts for 24 and 48 h is illustrated in Figure 4A. The treated cells exhibited distinctive morphological aberrations such as a shrunken appearance and cytoplasmic condensation when exposed to all the three at higher concentrations (50–200 µg/mL) [Figure 4b-d]. The IC_{50} concentration of the *A. mexicana* extracts required to inhibit the growth of HeLa cells after 24 and 48 h of exposure was 58.88 ± 2.74 and 38.01 ± 1.77 µg/mL, respectively, for the aqueous extract; 87.07 ± 4.65 and 57.54 ± 1.08 µg/mL, respectively, for the methanol extract; and 67.60 ± 2.47 and 38.90 ± 2.35 µg/mL, respectively, for the ethanol extract. The percentage viability of MCF-7 cells exposed to the aqueous, methanol, and ethanol extracts for 24 and 48 h is illustrated in Figure 5A-D. The IC_{50} concentration of the

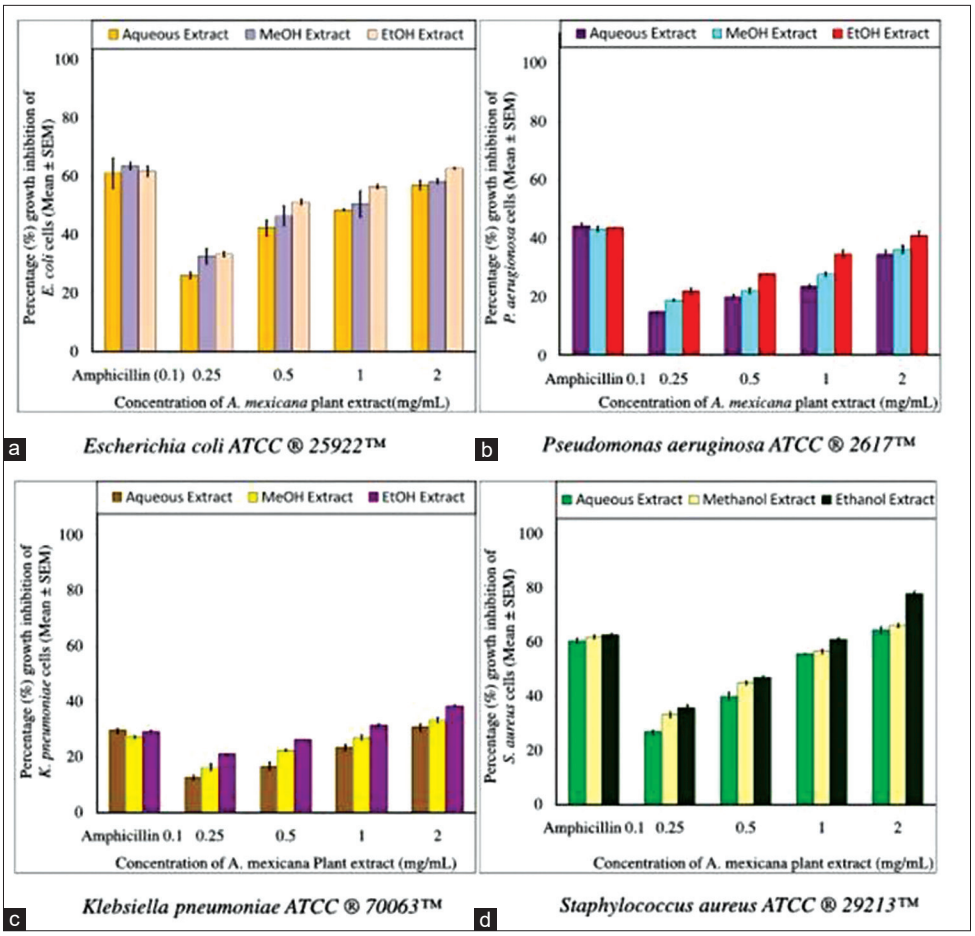

Figure 3: Representative histogram showing bacterial growth inhibition of (a) *Escherichia coli ATCC® 25922™* (b) *Pseudomonas aeruginosa ATCC® 2617™* (c) *Klebsiella pneumoniae ATCC® 70063™* and (d) *Staphylococcus aureus ATCC® 29213™* exposed to different concentrations (0.025, 0.05, 0.1 and 0.2 mg/mL) of aqueous, methanol and ethanol extracts of *Argemone mexicana* for 24 h. The results represent the means of three independent experiments, and error bars represent the standard error of the mean

A. mexicana extract required to inhibit the growth of MCF -7 cells after 24 and 48 h of exposure was 134.89 ± 2.36 and 95.50 ± 3.69 µg/mL, respectively, for the aqueous extract; 61.66 ± 3.87 and 58.88 ± 4.16 µg/mL, respectively, for the methanol extract; and 99.72 ± 2.52 and 64.56 ± 1.18 µg/mL, respectively, for the ethanol extract. The viability of HCT -15 cells exposed to 10–200 µg/mL of the three solvent extracts is illustrated in Figure 6A-D. The IC_{50} concentrations of the aqueous, methanol and ethanol extracts required to inhibit the growth of HCT-15 cells after 24 and 48 h of exposure were 60.25 ± 2.60 and 56.23 ± 3.98 µg/mL, 58.58 ± 4.66 and 41.68 ± 2.16 µg/mL, and 46.77 ± 2.66 and 24.54 ± 1.58 µg/mL, respectively. Thus, 93.68% ± 0.99% of the HCT-15 cells were killed when exposed to 200 µg/mL of the ethanol extract for 24 h, and 96.35% ± 0.53% of the HCT-15 cells were killed after a 48-h exposure to the ethanol extract.

The genotoxic effect on HeLa, MCF-7, and HCT-15 cells exposed to increasing concentrations of the aqueous, methanol, and ethanol *A. mexicana* plant extracts is illustrated in Figure 7a-c. On assessing the genotoxic effects of *A. mexicana* through DNA fragmentation in cells exposed to different concentrations (10–200 µg/mL) of the three extracts, 50–200 µg/mL methanol and ethanol extracts were found to be significantly genotoxic to HeLa and MCF-7 cells, causing more DNA fragmentation. Significant DNA fragmentation was observed in the cells treated with the 50–200 µg/mL plant extract; a characteristic ladder pattern was observed on the agarose gel [Figure 7a-c, Lanes 5–7]. By contrast, minimum or no DNA fragmentation was observed in control cells and HeLa and MCF-7 cells treated with a lower concentration of the extracts (10–25 µg/mL) [Figure 7a-c, Lanes 2–4].

Discussion

In the present study, we screened the phytochemical content and determined the antioxidant, antimicrobial, and cytotoxic potentials of different solvent extracts of *A. mexicana.* The phytochemical screening revealed a mixture of phytochemicals, including phenolics, flavonoids, and tannins. Phytochemicals screened from aqueous, methanol, and ethanol extracts showed higher amounts of phenolic content, followed by flavonoid and tannin contents. The ethanol extract was found to be rich in the total phenol content compared with the methanol and aqueous extracts. Medicinal plants possess secondary metabolites such as phenolic compounds that have many biological properties, including antioxidant and antimicrobial properties.[2,17] Numerous studies have been conducted on the free radical scavenging properties of phytoconstituents of various medicinal plants. The biological potential, including the free radical scavenging activity, of different solvent extracts of *A. mexicana* was determined through various antioxidant assays. In the present study, ABTS and DPPH radical scavenging assays confirmed the strong free radical scavenging activity of *A. mexicana*, wherein an increase in activity was observed with an increase in plant extract concentration from 10 to 200 µg/mL, similar to documented reports.[18,19] The reducing power of extracts serves as a significant indicator of

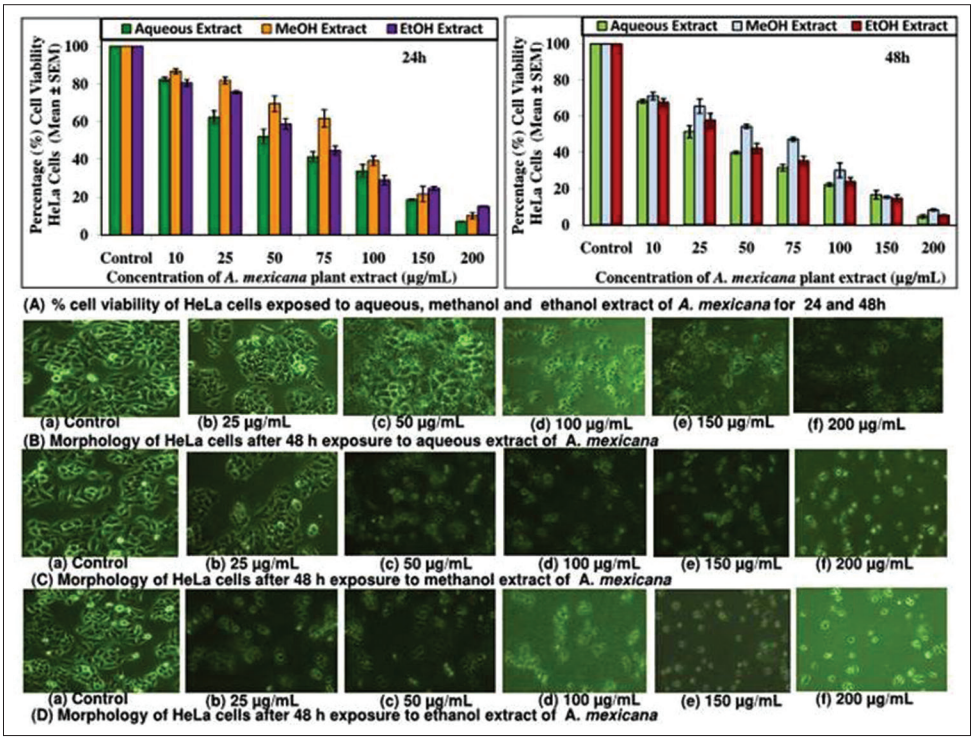

Figure 4: (A) Representative histogram showing *in vitro* dose-dependent cytotoxicity of aqueous, methanol, and ethanol extract of *Argemone mexicana* of on HeLa cells exposed for 24 h and 48 h. Cell morphology of HeLa cells after 48 h exposure to (B) aqueous, (C) methanol, and (D) ethanol extracts of *Argemone mexicana* plant at different concentrations. All images are taken at ×20 with Carl Zeiss phase-contrast microscope

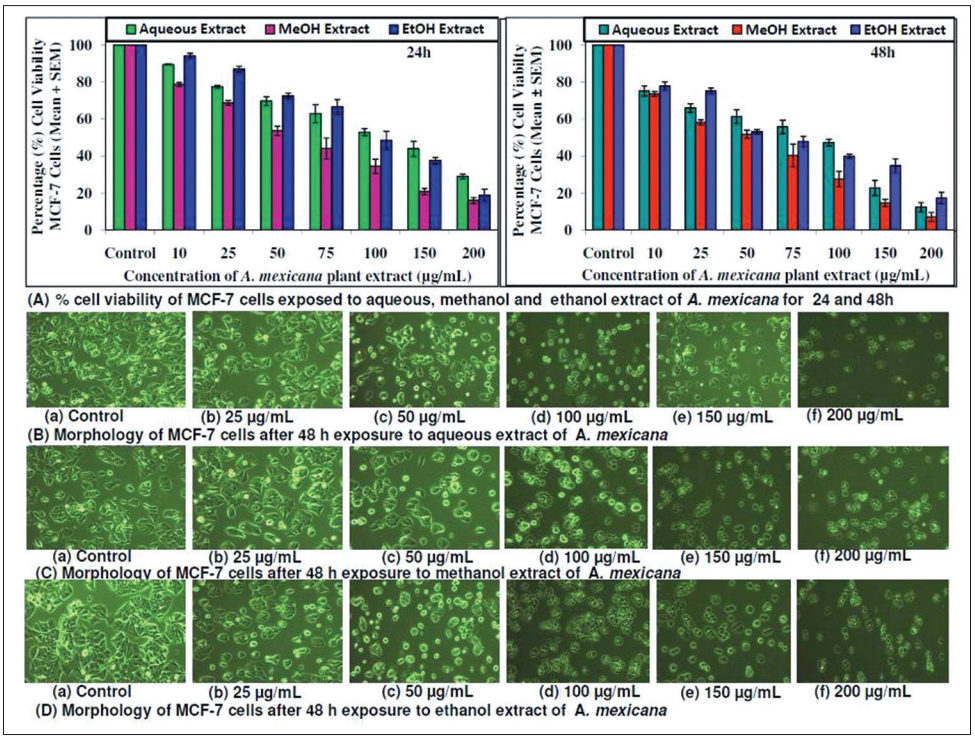

Figure 5: (A) Representative histogram showing *in vitro* dose-dependent cytotoxicity of aqueous, methanol and ethanol extract of *Argemone mexicana* of on MCF-7 cells exposed for 24 h and 48 h. Cell morphology of MCF-7 cells after 48 h exposure to (B) aqueous, (C) methanol, and (D) ethanol extracts of *Argemone mexicana* plant at different concentrations. All images are taken at ×20 with Carl Zeiss phase-contrast microscope

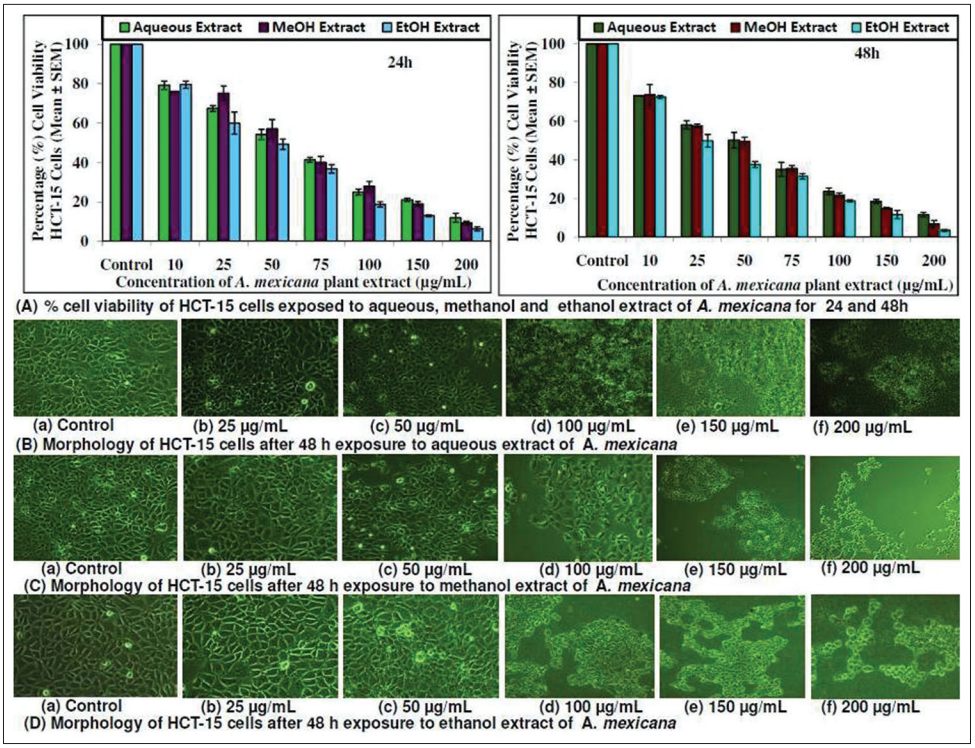

Figure 6: (A) Representative histogram showing *in vitro* dose-dependent cytotoxicity of aqueous, methanol, and ethanol extract of *Argemone mexicana* of on HCT-15 cells exposed for 24 h and 48 h. Cell morphology of HCT-15 cells after 48 h exposure to (B) aqueous, (C) methanol and (D) ethanol extracts of *Argemone mexicana* plant at different concentrations. All images are taken at ×20 with the Carl Zeiss phase-contrast microscope

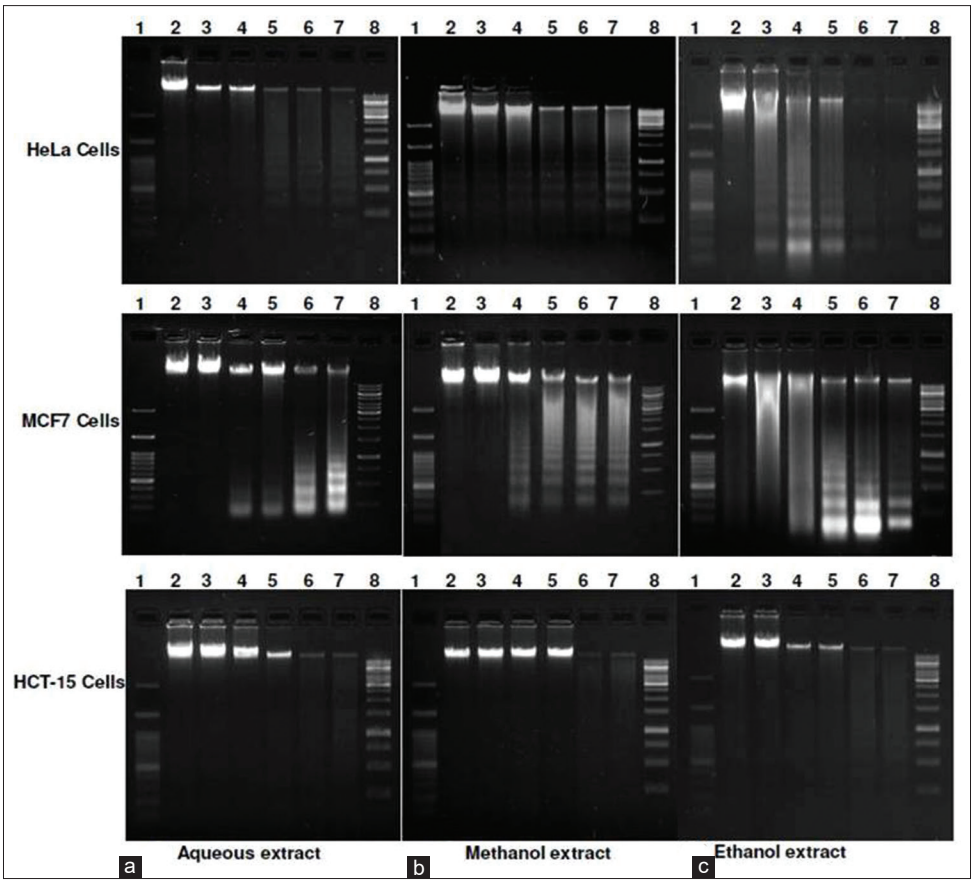

Figure 7: Representative agarose gel images showing DNA fragmentation in HeLa cells (upper panel), MCF-7 cells (middle panel) and HCT-15 cells (lower panel) treated with (a) aqueous, (b) methanol and (c) ethanol extracts of *Argemone mexicana* at lower to higher concentrations. In each representative gel, lane 1 is 100 bp DNA marker: Lane 2 is DNA from control cells followed by lane 3, 4, 5, 6, and 7 are DNA from 10, 25, 50, 100 and 200 μg/mL plant extract treated cells respectively, and lane 8: Is 1 kb DNA marker

the antioxidant activity where the reducing power increases with an increase in the plant extract concentration. The present study demonstrated an increased, reducing power of both the aqueous and ethanol extracts of *A. mexicana*.

The antimicrobial property of medicinal plants has been reported all over the world.[17,20] This study was conducted to evaluate the antibacterial potential of *A. mexicana* plant extracts against human pathogenic bacteria. Among tested *A. mexicana* samples, the ethanol extract exhibited the maximum growth inhibition of *E. coli* and *S. aureus* at a concentration of 2 mg/mL. Previous studies have reported that the methanol extract of *A. mexicana* exhibits the maximum activity at 100 mg/mL against *E. coli* and *S. aureus*.[21] However, in the present study, a substantially lower concentration (2 mg/mL) was required to inhibit the maximum growth of *E. coli* and *S. aureus*. On testing the aqueous, methanol, and ethanol extracts of the whole plant through antimicrobial assays, varying degrees of antimicrobial activity were observed against all the tested Gram-positive and Gram-negative bacteria, with the ethanol extract of *A. mexicana* exhibiting the greatest activity against the tested bacterial strains.

To explore the cytotoxic and genotoxic potential of the crude *A. mexicana* extract, HeLa, MCF-7, and HCT15 cancer cell lines were exposed to the aqueous, methanol, and ethanol extracts. All three tested extracts showed similar but slightly differing results, with the ethanol extract inducing the maximum growth inhibition of all the tested cells at 50–200 μg/mL. The analysis of cytotoxicity of different solvent extracts of *A. mexicana* revealed significantly higher cytotoxic activity of the ethanol extract than of the aqueous and methanol extracts. Previous studies on the cytotoxicity of *A. mexicana* stem and leaves have reported maximum inhibitory activity at 300 μg/mL.[14,15] By using the DNA fragmentation assay, we confirmed the genotoxic effects of different concentrations of the aqueous, methanol, and ethanol plant extracts on cancer cell lines exposed to. Extensive DNA double-strand breaks were observed in the HeLa, MCF7, and HCT-15 cells exposed to plant extracts, demonstrating the powerful genotoxic potential of *A. mexicana.* Thus, the present study provides extensive evidence supporting the cytotoxic and apoptotic potential of the *A. mexicana* plant.

Conclusion

The crude extract of the *A. mexicana* plant exhibited potent antioxidant, antibacterial, and cytotoxic activities comparatively higher than those reported by earlier studies.

These biological activities may be due to the presence of relevant secondary metabolites such as phenolics, flavonoids, and tannins in the plant species.

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Conflicts of interest

There are no conflicts of interest.

References

1. Karagoz A, Artun FT, Ozcan G, Melikoglu G, Anil S, Kultur S, Sutlupinar N. *In vitro* evaluation of antioxidant activity of some plant methanol extracts. Biotechnol Biotechnol Equip 2015;29:1184-9.
2. Ayoub Z, Mehta A. Medicinal plants as potential source of antioxidant agents: A review. Asian J Pharm Clin Res 2018;11:50-6.
3. Arya P, Mehta JP. A review on antimicrobial and Himalayan medicinal plants. Environ Conserv J 2017;18:46-62.
4. Mahato TK, Sharma K. Study of medicinal herbs and its antibacterial activity: A review. J Drug Deliv Ther 2018;8:47-54.
5. Bailon-Moscoso N, Romero-Benavides JC, Tinitana-Imaicela F, Ostrosky-Wegman P. Medicinal plants of Ecuador: A review of plants with anticancer potential and their chemical composition. Medicinal Chemistry Research 2015;24:2283-96.
6. Shukla S, Mehta A. Anticancer potential of medicinal plants and their phytochemicals: A review. Braz J Botany 2015;38:199-210.
7. Husna SA, Reddy VJ. A review on *Argemone mexicana*. Int J Pharmacologuical Res 2017;7:170-4.
8. Tewari D, Mocan A, Parvanov ED, Sah AN, Nabavi SM, Huminiecki L, *et al*. Ethnopharmacological Approaches for therapy of Jaundice: Part I. Frontiers in Pharmacology 2017;8:518.
9. Sharanappa R, Vidyasagar GM. Plant profile, phytochemistry and pharmacology of *Argemone mexicana*. Linn. A review. Int J Pharm Pharma Sci 2014;6:45-53.
10. Dutta S, Hazara K, Paria D, Ghoshal S, Dixit AK, Rao MM. Phyto pharmacognostical evaluation of *Argemone mexicana* L. Seeds. J Pharmacogn Phytochem 2018;7:1507-13.
11. Magaji MB, Haruna Y, Muhammad A. Phytochemical screening and antioxidant activity of *Argemone mexicana* leaf methanol extract. Ann Pharmacol Pharma 2019;4:1167-72.
12. Andleeb S, Alsalme A, Al-Zaqri N, Warad I, Alkahtani J, Bukhari SM. *In-vitro* antibacterial and antifungal properties of the organic solvent extract of *Argemone mexicana* L. Journal of King Saud University – Science 2020;32:2053-8.
13. Al Nasar IS, Koko WS, Khan TA, Elghazali GE. *In vitro* antitoxoplasmal activity of some medicinal plants. Pharmacol Mag 2019;15:568-72.
14. More NV, Kharat AS. Antifungal and anticancer potential of *Argemone mexicana* L. Medicines (Basel) 2016;3:28.
15. Prabhakaran D, Senthamilselvi MM, Rajeshkanna A. Anticancer activity of *Argemone mexicana* (Flowers) against human liver cancer (HepG2) cell line. Asian J Pharma Clin Res 2017;10:351-3.
16. Durgawale PP, Patil MN, Joshi SA, Korabu KS, Datkhile KD. Studies on phytoconstituents, *in vitro* antioxidant, antibacterial, antiparasitic, antimicrobial, and anticancer potential of medicinal plant *Lasiosiphon eriocephalus* decne (Family: Thymelaeaceae). J Natural Sci Biol Med 2019;10:38-47.
17. Khameneh B, Iranshahy M, Soheili V, Fazly Bazzaz BS. Review on plant antimicrobials: A mechanistic viewpoint. Antimicrob Resist Infect Control 2019;8:118.
18. Zahin M, Ahmad I, Shireen I, Husain FM, Aqil F. Broad spectrum antioxidant properties of 20 Indian medicinal plants. J Herbs Spices Med Plants 2016;22:118-29.
19. De P, Das S, Mukhopadhyay MJ. Total antioxidant profile of some common medicinal plants of West Bengal. Int J Pharmacogn Pharma Res 2018;10:240-5.
20. Adnan M, Ali S, Sheikh K, Amber R. Review on antibacterial activity of Himalayan medicinal plants traditionally used to treat pneumonia and tuberculosis. J Pharm Pharmacol 2019;71:1599-625.
21. Haruna Y, Ukamaka O. Anti-Microbial and Anti-Fungal Activities of Methanol Extract of Argemone mexicana and its Potential Anti-Hepatitis Promises. Journal of Clinical & Experimental Pharmacology 2018;8:251.