International Journal of Herbal Medicine
Available online at www.florajournal.com

E-ISSN: 2321-2187
P-ISSN: 2394-0514
www.florajournal.com
IJHM 2020; 8(3): 36-38
Received: 19-03-2020
Accepted: 21-04-2020

Sivakumar Eniya
Department of Biochemistry, PSG College of Arts & Science, Coimbatore, Tamil Nadu, India

Thangavel Thirunavukkarsu
Department of Biochemistry, PSG College of Arts & Science, Coimbatore, Tamil Nadu, India

S Mohandass
Department of Biochemistry, PSG College of Arts & Science, Coimbatore, Tamil Nadu, India

Corresponding Author:
S Mohandass
Department of Biochemistry, PSG College of Arts & Science, Coimbatore, Tamil Nadu, India

Antiproliferative efficacy of sorghum bran extracts against MCF-7 breast cancer cell lines

Sivakumar Eniya, Thangavel Thirunavukkarsu and S Mohandass

Abstract
Over the past decade, there has been an increasing interest in using nanotechnology for cancer therapy. The development of smart targeted nanoparticles (NPs) that can deliver drugs at a sustained rate directly to cancer cells may provide better efficacy and lower toxicity for treating primary and advanced metastatic tumors. The present study was aimed to investigate the in vitro cytotoxicity effect of biogenic Zinc nanoparticles ZnONPs) against human breast cancer (MCF-7) cells towards the development of anticancer agent. Nanobiocomposite of zinc oxide nanoparticles conjugated with Sorghum Bran was produced and was confirmed using maximum UV–Vis absorption at 340 nm in the present work. The anti-cancerous nature of the synthesized zinc oxide conjugated nanobiocomposite on MCF-7 cell line was studied using MTT assay. The viability of the MCF-7 cells was decreased to 48% when it was treated with Sorghum bran conjugated zinc oxide nanobiocomposite. There was an immediate induction of cellular damage in terms of loss of cell membrane integrity, oxidative stress and apoptosis were found in the cell which treated with ZnONPs. This may be a first report on anti-MCF-7 property of biogenic ZnONPs, but mechanism needs to be elucidated.

Keywords: Sorghum, MTT assay, anticancer

1. Introduction
Medicinal plants and its herbal therapy have their important role in the treatment of various diseases. Besides their therapeutic potency, some of the medicinal plant has toxicity effect particularly in children and elderly people. So it is believed that medicinal plants are the natural products which are safe to consume and also less toxic. Traditional medicines promote the health maintenance and diagnose the diseases. The current research behind the medicinal plant therapy includes the mind, body and energy based healing, herbal remedies [1]. Ethanopharmacological screening of medicinal plants is considered to be an important approach where several medicinal plants have been reported as nontropic that altercates mental and cognitive functions by influencing various different pathway of brain [2]. In latest advancement, the medicinal plants are replaced by modern drugs. Many people used to consume the scientifically developed tablets and pills. Medicinal plant usage creates the recent awareness among the people to treat diseases and drug that is developed from bark, seed, fruit and other parts of body. It also acknowledged that their active action and it has been included in modern pharmacotherapy [3]. Secondary metabolites of medicinal plants play an important role in diagnosis of disease and this in turn is used for production of novel medicines. Large number of plants possesses many other activities such as antioxidant, anti-inflammatory, antibiotic, anti insecticidal. Those are widely used by the people all around the world. Evaluating the active plant derived compound, herbal drugs can help in the healthcare system to treat human diseases in future [4]. Sorghum is a world's fifth largest produced cereal after maize, rice, wheat and barley. Sorghum is also rich in bioactive compounds particularly 3-deoxyanthocyanins,tannins, phenolic acids and minerals such as calcium, phosphorous, iron, zinc and magnesium ,unsaturated lipids and complex B vitamins. Sorghum bran, that is usually discarded s waste, is identified to be rich in phenolic compounds.contains a phenolic compounds in the form of phenolic acids, flavonoids and condensed tannins which have antioxidant activity [5]. Anthocyanins are the poly phenolic pigments that belongs to a flavonoid group and it is responsible for the red-orange to blue-violet colours present in plant organs. They play relevant roles in plant propagation and ecophysiology and plant defense mechanisms and are responsible for the colour of fruits and vegetables. A large number of novel anthocyanin structures have been identified, including new families such as pyranoanthocyanins or anthocyanin oligomers; their biosynthesis pathways have been elucidated [6]. Furthermore, evidence about their benefits in human health has accumulated, and processes of anthocyanin absorption and biotransformation in the human organism have

started to be ascertained. Earlier, anthocyanin is only known for the colouring properties and now anthocyanin plays an important role in the health benefits as dietary antioxidants by preventing neuronal diseases, cancer ,diabetes, cardiovascular illness, inflammation and many other diseases [7]. The present study focuses on the isolation of anthocyanins from Sorghum bran, convert to a ZnO nanoparticle and evaluate its efficacy as anticancer in *in vitro* models.

2. Methodology

2.1 Preparation of Extract of the Sample

The Sorghum plant was collected from farming land in Coimbatore. The grains in the plants are removed and the bran is separated from the plant. The obtained bran was powdered. 10g of bran powder were taken in a conical flask for sample extraction by using 100ml of chloroform solvent for 72hrs.After the incubation, the extract was air dried. The crude that was obtained was dissolved in water and used for studies.

2.2 Synthesis of Zinc Oxide Nanoparticles

In a typical reaction mixture, 5-10 ml of the aqueous extract of red sorghumbran was added to 300 ml of 4Mm of aqueous zinc sulphate heptahydrate solution and stirred at room temperature for 5 min s to achieve the pale yellow solution. After that, 1M sodium hydroxide solution is added to the mixture drop by drop with continuous stirring at room temperature. The yellow colour of the above mixture starts change to yellowish suspension at pH 12. The suspended particles were purified by dispensing in sterile distilled water and centrifuged thrice. Further, the white precipitate particles were washed with ethanol to remove the impurities for the final product. The white powder was obtained after drying at 60ºC in vaccum oven for 6 hrs.The formed nanocrytals were confirmed by its UV Vis absorption at 340nms.

2.3 In *vitro* screening of Red Sorghum branZnOPs on MCF-7 Cancer cell line.by MTT Assay [8]

2.3.1 Cell line and culture

MCF 7 cell line was obtained from National Centre for Cell Sciences, Pune (NCCS). The cells were maintained in DMEM supplemented with 10% FBS, penicillin (100 U/ml), and streptomycin (100 µg/ml) in a humidified atmosphere of 50 µg/ml CO_2 at 37 °C.

2.3.2 Procedure

Cells (1 × 10^5/well) were plated in 24-well plates and incubated in 37^0C with 5% CO_2 condition. After the cell reaches the confluence, the various concentrations of the samples were added and incubated for 24hrs. After incubation, the sample was removed from the well and washed with phosphate-buffered saline (pH 7.4) or DMEM without serum. 100µl/well (5mg/ml) of 0.5% 3-(4,5-dimethyl-2-thiazolyl)-2,5-diphenyl--tetrazolium bromide (MTT) was added and incubated for 4 hours. After incubation, 1ml of DMSO was added in all the wells. The absorbance at 570nm was measured with UV- Spectrophotometer using DMSO as the blank. Measurements were performed and the concentration required for a 50% inhibition (IC50) was determined graphically. The % cell viability was calculated using the following formula:

% Cell viability = A570 of treated cells / A570 of control cells × 100

Graphs are plotted using the % of Cell Viability at Y-axis and concentration of the sample in X-axis. Cell control and sample control is included in each assay to compare the full cell viability assessment.

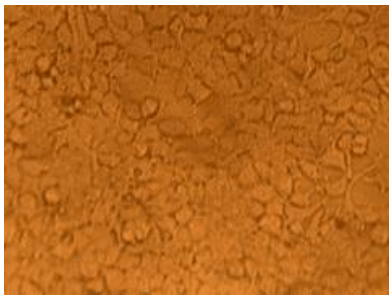
3. Results and Discussion

Pharmacological studies fused with the many clinical observations and their effects of chronic administration of chemotherapy agent for the cancer the cancer treatment. Anticancer drugs cause an irreversible pharmacological response, where it requires long observation times and it is not measurable accurately. The empirical approach to cancer chemotherapy has often been successful [9]. Dietary antioxidants and various phytochemicals occurring in the herbs to prevent cancer that induces oxidative damages. Oxidative stress is a component of environmental toxicity during the cancer process. ROS potentially play an important role in the cancer. The level of ROS determines the risk of cancer in humans. Low level of ROS can be beneficial in the normal function, whereas there is the increased risk of cancer if occurs the chronic exposure to ROS. Increased level of ROS tends to induce apoptosis and cell death in various types of cancer. The antioxidant activity of phytochemicals is determined and used to prevent cancer [10]. Results obtained from the previous phases prove that, nanoparticles of red sorghum bran (ZnONPs) was found to exert better free radical scavenging, antioxidant effects in all the *in vitro* models.

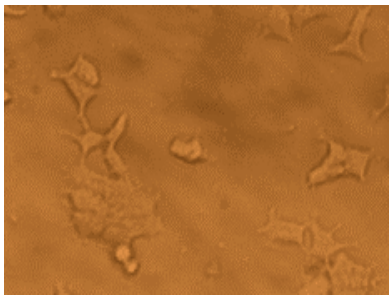
3.1 MTT ASSAY

The MTT (3-[4,5-dimethylthiazol-2-yl]-2,5 diphenyltetrazolium bromide) assay is based on the conversion of MTT into formazan crystals by living cells, which determines mitochondrial activity. Since for most cell populations the total mitochondrial activity is related to the number of viable cells, this assay is broadly used to measure the in vitro cytotoxic effects of drugs on cell lines or primary patient cells.MTT assay allowing one to defect cell stress upon exposure to a toxic agent in the absence of direct cell death [11].

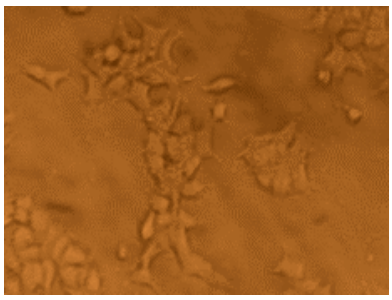
To determine the anticancer effect of ZnONP's of red sorghum bran, MCF7 cell lines were used. Effect of synthesized nanoparticles on cell response of the Human breast cancer (MCF-7) cell line by using UV-vis spectrophotometer MTT assay. When the MCF7 cell lines were exposed to different concentration of ZnONP's of red sorghum bran (7.8 µg/ml to 1000 µg/ml, the cell viability of the line decreased from 48.60% to 07.08%. The cell control have the absorbance of optical density at 0.395 and cell viability of 100%. IC_{50} value is found to be 7.8µg/ml [12].



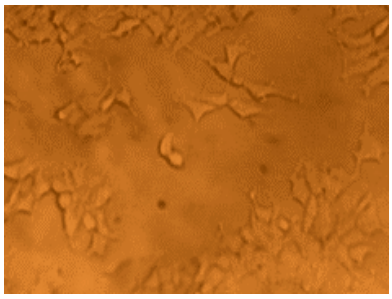
Normal cell line



1000µg/ml



62.5µg/ml



7.8µg/ml

Table 1: Shows the anticancer effect of ZnONP's of red sorghum bran on MCF7 cell line

S. No	Concentration (µg/ml)	Dilutions	Absorbance (O.D)	Cell viability%
1	1000	Neat	0.028	07.08
2	500	1:1	0.048	12.15
3	250	1:2	0.071	17.97
4	125	1:4	0.093	23.54
5	62.5	1:8	0.116	29.36
6	31.2	1:16	0.140	35.44
7	15.6	1.32	0.163	41.26
8	7.8	1.64	0.192	48.60
9	Cell control	-	0.395	100

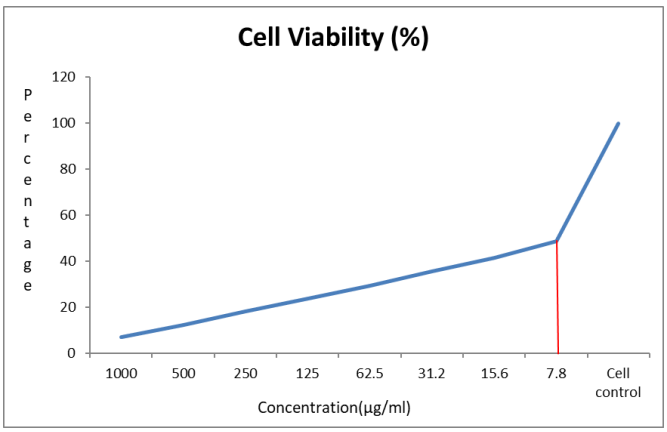


Fig 1: Represents the cell viability in %

It is reported in previous study that Pp-ZnO NPs also effectively inhibited the biofilm formation of *C. albicans* at 50 µg ml^{-1}. Cytotoxicity studies revealed that a single treatment with Pp-ZnO NPs significantly reduced the cell viability of breast cancer cell line MCF-7 cells at doses higher than 50 µg ml^{-1} [13, 14].

4. Conclusion

The *in vitro* antiproliferative and antiapoptotic activity of red sorghum bran ZnONPs were assessed against breast cancer cell line. The cytotoxic effect induced by the ZnONPs were also compared with control cells. The results obtained for viability and cytotoxicity assays showed that the exposure of cancer cells to red sorghum bran caused a steep decrease in the viability of the cells. The antiproliferative effect of red sorghm bran ZnONPs were found to be dose dependent in the cell line. The present study highlights the efficacy of Sorghum bran as antiproloferative, but the exact molecular mechanism needs to br elucidated.

5. References

1. Reena R, Afton C. Medicinal plants with traditional use: Ethnobotany in the Indian subcontinent. Clinics of Dermatology. 2018; 36(3):306-309.
2. Swati Vyas SL, Kothari, Sumita K. Nootropic Medicinal Plants: Therapeutic Alternatives for Alzheimer's disease. J Herbal Medicine. 2019; 100291: 17 -18.
3. Nima, Hui TM, Mandal P, Kalita A, Das K. An ethnobotanical study of traditional anti-inflammatory plants used by the Lohit community of Arunachal Pradesh, India. J Ethnopharmacology 2009; 125(2):234-245.
4. Fatemeh JK, Zahra L, Hossein AK. Medicinal Plants: Past History and Future Perspective. J Herbal Medicine Pharmacology. 2018; 7(1):1-7.
5. Awika LW, Rooney XW, Ronald LP, Luis CZ. Screening Methods to Measure Antioxidant Activity of Sorghum (Sorghum bicolor) and Sorghum Products. J Agriculrure Food Chemistry. 2003; 51:23:6657- 62.
6. Jin-Ming K, Lian-SC, Ngoh K, Tet FC, Brouillard R. Analysis and biological activities of anthocyanins. Phytochemistry. 2003; 64(5):923-933.
7. Castañeda-Ovando A, Pacheco-Hernández M, Páez-Hernández M, Rodríguez JA, Galán-VC. Chemical studies of anthocyanins: A review. Food Chemistry. 2009; 113(4):859-871.
8. Johan van Meerloo Gertjan JL, Kaspers JC. Cell Sensitivity Assays: The MTT Assay. Cancer Cell Culture 2011; 731:237-245.
9. Anna AP, Pavel AL. Precision Medicine in Oncology:In Vitro Drug Sensitivity and Resistance Test (DSRT) for Selection of Personalized Anticancer Therapy. Advanced Therapeutics 2020; 3:2- 7.
10. Jeyaraj M, Sathishkumar G, Sivanandhana G, Mubarak Ali M, Rajesh R, Arun G *et al*. Biogenic silver nanoparticles for cancer treatment: An experimental report. Colloids and Surfaces B: Biointerfaces 2013; 106:86-92.
11. Kristin B, Peter N, Rolf L, Claes RA. Predictive Value of Ex Vivo Chemosensitivity Assays for Individualized Cancer Chemotherapy: A Meta-Analysis. SLAS TECHNOLOGY: Translating Life Sciences Innovation. 2017; 22(3):306-314.
12. Mohammed JA, Maqusood A, Sudhir Kumar, Majeed Khan MA, Javed AS, Alrokayan A. Zinc oxide nanoparticles selectively induce apoptosis in human cancer cells through reactive oxygen species. International J Nanomedicine. 2012; 7: 845–857.
13. Balasubramanian M, Baskaralingam V, Sekar V, Karuppiah P, Mohamed Ali R, Kadarkarai M, *et al*. Biological therapeutics of *Pongamia pinnata* coated zinc oxide nanoparticles against clinically important pathogenic bacteria, fungi and MCF-7 breast cancer cells. Microbial Pathogenesis 2017; 104:268-277.
14. Huang CH, Sutapa B, Gaurav S, Sandwip KD, Kaushal R. Inorganic nanoparticles for cancer imaging and therapy. J Controlled Release. 2011; 155(3):344-357