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Homeopathic Silicea Exhibits Dose-Dependent Cytotoxicity In Mcf-7 Breast Cancer Cell Line: An In Vitro Study.

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Abstract: **Introduction**: Silicea is a widely used homoeopathic remedy which is known for supporting the body's natural healing processes, especially in chronic infections, skin diseases, and connective tissue weakness. Scientific evidence regarding its impact in cancer research is still scarce despite its therapeutic application. **Methods**: This study aims to evaluate the cytotoxic potential of Silicea on MCF-7 human breast cancer cell lines at potencies of 30CH, 200C, 1M and 10M. Using the MTT assay, metabolic activity and cell viability were assessed after treatment.**Result**: The results shows notable morphological changes such as cell shrinkage and loss of adherence, indicating reduced viability and possible apoptotic effects. These results indicate that Silicea may possess cytotoxic effects against breast cancer cells and should be explored in more detail through further research.

Keywords: Cytotoxicity, MCF-7 cell line, Homoeopathy.

I. INTRODUCTION

Breast cancer is the most commonly diagnosed malignancy in women worldwide and remains the second leading cause of cancer-related mortality(6). It arises from the uncontrolled growth of malignant cells within breast tissue, driven by a complex interplay of genetic, hormonal, and environmental factors. Genetic predispositions such as mutations in BRCA1, BRCA2, and P53 significantly increase susceptibility, particularly in individuals with a family history of breast or ovarian cancer (2). Although incidence rates are higher in developed nations, mortality is disproportionately greater in developing regions due to late diagnosis, limited healthcare access, and lack of awareness (1). As breast cancer develops through multifactorial mechanisms involving DNA damage, hormonal influences, and environmental exposures, understanding its biological basis is crucial for designing effective therapeutic strategies. Advances in research rely heavily on reliable in vitro models, with the MTT assay serving as a widely used colorimetric

technique for assessing cell viability and cytotoxicity by measuring mitochondrial activity[3]. This assay enables quantitative evaluation of anticancer agents and supports the development of improved therapeutic approaches. In this study, the cytotoxic effects of the well-known homoeopathic remedy Silicea at varying potencies (30CH, 200C, 1M, and 10M) on MCF-7 human breast cancer cell lines were evaluated using the MTT test[4]. The results demonstrate a decline in cell viability with increase in potency of the homeopathic medicine Silicea[5], highlighting its potential relevance in exploring alternative or complementary therapeutic strategies for breast cancer.

Principle of MTT Assay

The MTT assay is a widely used colorimetric test that measures cellular metabolic activity as a marker of cell viability, proliferation, and cytotoxicity. It relies on the reduction of the yellow tetrazolium salt MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) into insoluble purple formazan crystals by NAD(P)H-dependent oxidoreductase enzymes in viable cells[7,8].

Once formed, the formazan is solubilized—commonly in DMSO or acidified solvents—and quantified by measuring absorbance at 570 nm using a spectrophotometer or ELISA plate reader[12]. The amount of formazan produced is directly proportional to the number of metabolically active cells; therefore, a reduction in formazan formation indicates decreased cell viability or cytotoxic effects[8]. However, the MTT assay has certain limitations: increased mitochondrial mass or metabolic hyperactivation may artificially elevate formazan levels without reflecting true cell number[9,11]. Additionally, some compounds can non-enzymatically reduce MTT, leading to falsely high viability readings[10].

II. MATERIALS AND METHODS:

Reagents and Chemicals:

- Dulbecco's Modified Eagle Medium
- Fetal Bovine Saline (FBS)
- 1% antibiotic solution containing Penicillin (100 U/mL)
- Streptomycin (0.1 mg/mL)
- Amphotericin B (0.25 µg/mL)
- *Silicea* potencies (test sample)

Equipment:

- Laminar Air Flow Unit: Beston Industries, Cat No: BLV-0255
- Phase Contrast Fluorescence Microscope: LABOMED, TCM 400
- CO2 Incubator: New Brunswick
- MultiSKAN Skyhigh Spectrophotometer: SkanIt Software6.1.1, ThermoScientific.

MTT Assay:

- **Methodology:**

The MCF-7 human breast adenocarcinoma cell line was obtained from a standard cell culture repository and maintained under aseptic conditions in Dulbecco's Modified Eagle Medium (DMEM) supplemented with 10% fetal bovine serum (FBS), 1% penicillin-streptomycin antibiotic solution, and 1% L-glutamine to support optimal cell growth. The cells were cultured in T-25 culture flasks and incubated at 37°C in a humidified atmosphere containing 5% CO_2 until reaching approximately 80–90% confluence. For the cytotoxicity assay, viable MCF-7 cells were trypsinized using 0.25% trypsin-EDTA, counted using a hemocytometer, and seeded at a density of 10,000 cells per well into a sterile 96-well tissue culture plate. The plates were then incubated for 24 hours to allow for adequate cell attachment. Following this period, the cells were treated with the homeopathic remedy Silicea at four different potencies—30CH, 200C, 1M, and 10M—by adding 20 µL of each potency to achieve a final volume of 200 µL per well. Wells containing untreated cells served as negative controls, while blank wells containing only medium without cells were used to correct for background absorbance. All treatments were performed in triplicate to ensure reproducibility and statistical reliability. After a 24-hour exposure period, the medium in each well was carefully aspirated and replaced with 100 µL of MTT solution (0.5 mg/mL in phosphate-buffered saline

[PBS]), followed by incubation for 2 hours at 37°C to allow viable cells to reduce the MTT reagent into insoluble formazan crystals. The supernatant was then gently removed without disturbing the crystal layer, and 100 µL of 100% dimethyl sulfoxide (DMSO) was added to each well to dissolve the formazan product completely. The optical density (OD) of the resulting purple-colored solution was measured at a wavelength of 570 nm using an ELISA plate reader (MultiSKAN SkyHigh, Thermo Scientific). The obtained absorbance values were used to calculate the percentage of cell viability relative to the untreated control group, thereby determining the cytotoxic potential of Silicea at various potencies on MCF-7 cells.

The healing response of Silicea was evaluated through cell viability, calculated using the following formula:

$$\textbf{Percentage of Cell Viability (\%)} = \frac{\textbf{Average absorbance of treated cells}}{\textbf{Average absorbance of control cells}} \times \mathbf{100}$$

III. STATISTICAL ANALYSIS

Statistical comparisons were made between treated and control groups to determine the significance of observed differences in cell viability. The dose-dependent reduction in viability was statistically significant ($p < 0.05$), confirming the efficacy of Silicea in inducing cytotoxic effects on MCF-7 cells.

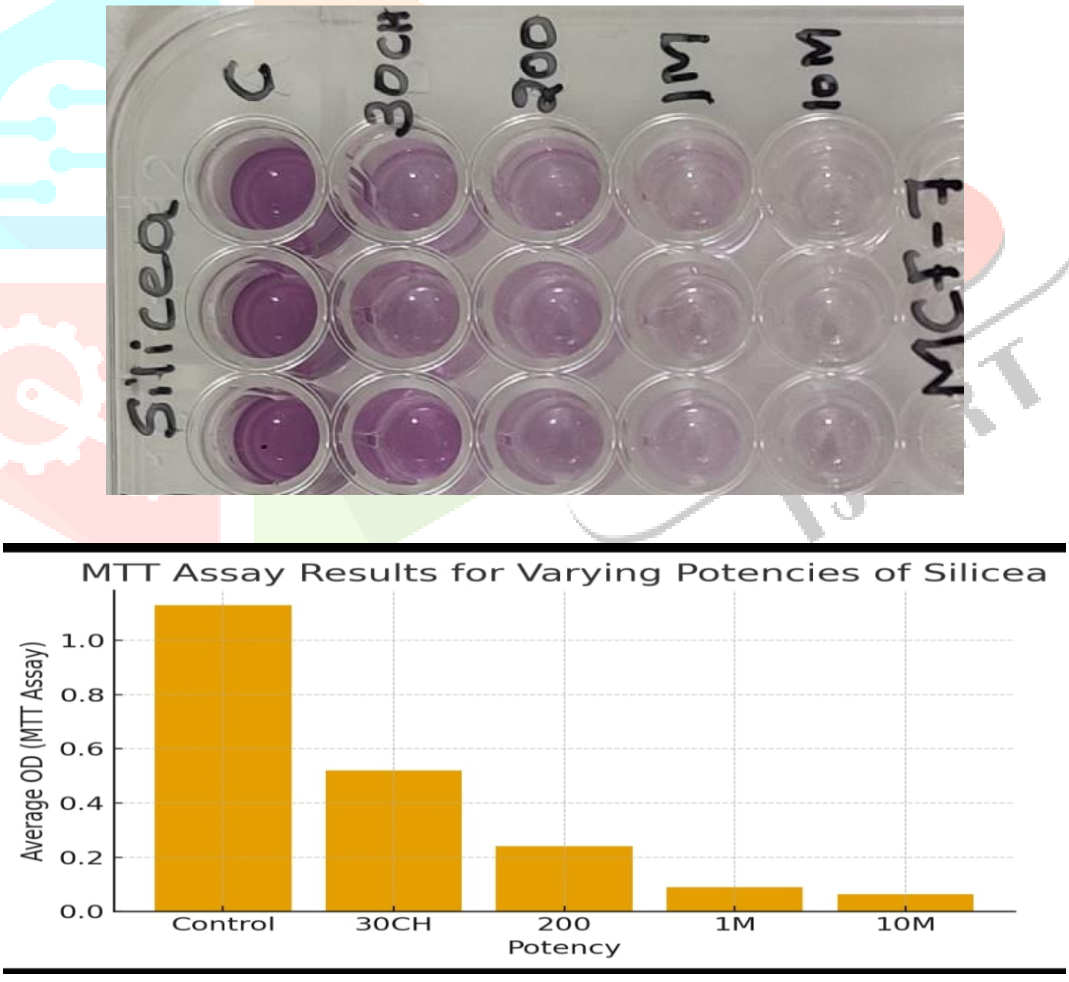

FIG 1: MTT Assay results

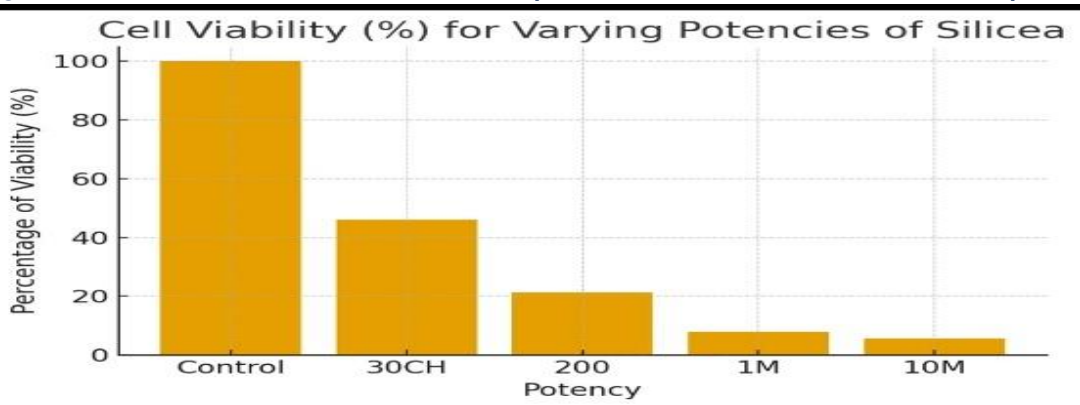

FIG 2: Cell Viability

RESULT: MTT Assay by different potencies of *Silicea*

Table 1: MTT Assay Results for Varying Potency of Silicea

Potency	OD1	OD2	OD3	Average OD
Control	1.130	1.120	1.140	1.130
30CH	0.528	0.515	0.518	0.520
200	0.244	0.242	0.237	0.241
1M	0.087	0.091	0.087	0.088
10M	0.064	0.067	0.062	0.064

Table 2: Percentage of Viability for Varying Potencies of Silicea

Potency	Percentage of Viability (%)
30CH	46.05
200	21.33
1M	7.82
10M	5.69

IV. DISCUSSION:

The findings of this study demonstrate that Silicea exhibits significant dose-dependent cytotoxic effects on MCF-7 human breast cancer cells, with the MTT assay revealing a progressive decrease in cell viability as potency increases, culminating in the most substantial reduction at 10M. The decrease in viability to 46.05% at 30CH indicates moderate cytotoxicity, while the reduction to 21.33% at 200C reflects a stronger effect. Higher potencies, including 1M and 10M, produced marked cytotoxicity with viability dropping to 7.82% and 5.69%, respectively, confirming that increased dilution and potentization enhance Silicea's inhibitory effect on cancer cell growth. Morphological observations further support these results, as lower potencies showed early apoptotic indicators such as cell shrinkage and membrane blebbing, whereas intermediate potencies displayed more prominent apoptotic features including nuclear fragmentation. At the highest potencies, extensive cell detachment and severe loss of cellular integrity were observed, consistent with pronounced apoptosis. These findings suggest that Silicea may hold potential as a complementary therapeutic agent in breast cancer management, though further studies are needed to identify its underlying mechanisms, evaluate its effects on normal cells, and assess its safety and therapeutic window. Future research should also investigate its possible synergistic role alongside conventional cancer therapies, explore its activity across different cancer cell lines, and validate the in vitro results through in vivo studies and clinical trials. Despite

its promising effects, the study is limited by its reliance on a single cell line and the absence of in vivo confirmation, underscoring the need for broader investigative approaches.

V. CONCULSION:

This study used the MTT assay to assess Silicea's cytotoxic effects on the human breast cancer cell line MCF-7. The findings showed a dose-dependent decrease in cell viability, suggesting that Silicea may be able to stop the proliferation of cancer cells. In summary, this study highlights that Silicea exhibits cytotoxic effects on MCF-7 breast cancer cells, significantly reducing their viability in a dose-dependent manner, and suggests its potential as a complementary therapeutic agent in breast cancer management.

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DISCLOSURE: No conflicts of interest.

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