# Medicine and Community Health Archives

https://mchapub.com/index.php/mcha

ISSN: 3023-3011

October, 2023|Volume 01|Issue 03| DOI: https://doi.org/10.23958/mcha/vol01/i03/33

**Clinical Case Reports** 



# The Safety of Glucosodiene on an In-Vitro Biopsy Cell Line Model.

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Received 05-09-2023| Accepted 14-10-2023| Published Online 19-10-2023

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### **Abstract**

This manuscript explores the safety and potential efficacy of glucosodiene, an isomeric alkaline glucose compound, as a therapeutic agent for cancer treatment. The study focuses on conducting safety tests using BJ1 cells (Normal Skin Fibroblasts) and evaluates the cytotoxic effects of glucosodiene on these cells. The results indicate that glucosodiene, at a concentration of 100 ppm, does not exhibit toxicity or impair normal cellular functions. These findings suggest that the compound may selectively target tumor cells without affecting healthy cells. However, further research is needed to understand the mechanisms of action and confirm its efficacy.

Keywords: Glucosodiene theory, Alkaline glucosodiene molecules, Safety of Glucosodiene.

#### Introduction:

Glucosodiene is an isomeric alkaline glucose compound formed by the reaction or heating of sodium bicarbonate with dextrose monohydrate. Its effectiveness has been documented in a theoretical framework targeting tumors through its metabolic activity [1,2]. Although research on the structural conformation of glucosodiene is ongoing [3,4], its safety and specific efficacy warrant further investigation. This manuscript focuses conducting safety tests on this novel compound using BJ1 cells (Normal Skin Fibroblasts), particularly after the emergence of a case report documenting the effectiveness of glucosodiene in a woman with triple-negative breast cancer (TNBC)

metastatic to the bones, leading to her recovery <sup>[5]</sup>. This necessitates additional documentation of reference dosages and safety results.

#### **Materials and Methods:**

#### **Chemicals**

Glucosodiene was prepared by adding 3.5 grams of dextrose monohydrate to 2.5 grams of sodium bicarbonate in 100 mL of distilled water and heating them to 100°C until the appearance of bubbles indicating sodium bicarbonate decomposition in water. This led to a substitution reaction between hydrogen and sodium atoms present in dextrose and sodium bicarbonate,

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respectively. The resulting material known as glucosodiene was then placed in a lyophilization apparatus, and a known quantity of the substance was obtained at a concentration of 100 ppm.

### Cell Line

BJ1 (Normal Skin Fibroblast) cells were obtained from the Bioassay-Cell Culture Laboratory at the

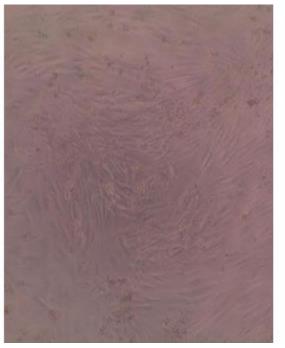
National Research Center, Dokki, Cairo, Egypt. Cytotoxic activity tests were conducted at the highest concentration of 100 ppm.

# **Results:**

Cytotoxic effect on human normal fibroblast cell line (BJ1)

Sample Code	<b>LC</b> <sub>50</sub> (μg/ml)	LC <sub>90</sub> (µg/ml)	Remarks
glucosodiene molecules			0.3% at 100ppm
DMSO			1% at 100ppm
Negative control			0 %

Figure 1







B Glucosodiene

Cell viability was assessed by the mitochondrial dependent reduction of yellow MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) to purple formazan <sup>[6]</sup>. Procedure: All the following procedures were done in a sterile area using a Laminar flow cabinet biosafety class II level (Baker, SG403INT, Sanford, ME, USA). Cells were suspended in DMEM-F12 medium, 1% antibiotic-antimycotic mixture (10,000U/ml Potassium Penicillin, 10,000µg/ml Streptomycin

Sulfate and 25µg/ml Amphotericin B) and 1% L-glutamine at 37 °C under 5% CO2. Cells were batch cultured for 10 days, then seeded at concentration of 10x10<sup>3</sup> cells/well in fresh complete growth medium in 96-well microtiter plastic plates at 37 °C for 24 h under 5% CO2 using a water jacketed Carbon dioxide incubator (Sheldon, TC2323, Cornelius, OR, USA).

Media was aspirated, fresh medium (without serum) was added and cells were incubated either

alone (negative control) or with different concentrations of sample to give a final concentration of (100-50-25-12.5-6.25-3.125-0.78 and 1.56 ug/ml). After 48 h of incubation, medium was aspirated, 40ul MTT salt (2.5µg/ml) were added to each well and incubated for further four hours at 37°C under 5% CO2. To stop the reaction and dissolving the formed crystals, 200µL of 10% Sodium dodecyl sulphate (SDS) in deionized water was added to each well and incubated overnight at 37°C. DOX were used as positive control at 100µg/ml gives 100% lethality under the same conditions [7]. The absorbance was then measured using a microplate multi-well reader (Bio-Rad Laboratories Inc., model 3350, Hercules, California, USA) at 595nm and a reference wavelength of 620nm. A statistical significance was tested between samples and negative control (cells with vehicle) using independent t-test by SPSS 11 program. DMSO is the vehicle used for dissolution of plant extracts and its final concentration on the cells was less than 0.3%. Figure 1 A,B] The percentage of change in viability was calculated according to the formula:

((Reading of extract / Reading of negative control) -1) x 100)

A probit analysis was carried for IC<sub>50</sub> and IC<sub>90</sub> determination using SPSS 11 program.

## **Discussion:**

The laboratory tests conducted on BJ1 normal skin fibroblast cell samples, replicated approximately 10 times, demonstrated the safety of glucosodiene on normal cells. At a concentration of 100 ppm, its toxicity was 0.3%, indicating its safety on normal cells. High doses of glucosodiene did not induce cellular toxicity or organic functional impairment, as observed in a case study involving a TNBC patient with bone metastasis who received glucosodiene treatment for 20 days. Laboratory results revealed that liver, kidney, heart, pancreas functions, blood parameters, blood pH, and urine analysis were all within the normal range [5]. This suggests that the new alkaline glucose isomer may disrupt or inhibit glucose metabolism within the tumor, known as the Warburg effect, which affects

cancer cells without affecting normal cells. However, further research is needed to confirm and understand the dynamics of glucosodiene's action as an alkaline glucose isomer.

#### **Conclusion:**

The safety tests conducted in the laboratory on BJ1 normal skin fibroblast cell samples indicate that glucosodiene at a concentration of 100 ppm does not exhibit toxicity or impair the normal cellular functions of these cells. These findings support the potential safety of glucosodiene for normal cells and provide a basis for further investigation into its efficacy and mechanism of action. Further research is warranted to explore the specific effects of glucosodiene on tumor cells and its potential as a therapeutic agent. Animal studies and clinical trials should be conducted to evaluate its safety and effectiveness in vivo. Additionally, elucidating the underlying mechanisms of glucosodiene's action on tumor cells, particularly its impact on glucose metabolism, would contribute to a better understanding of its potential as an anticancer agent. Overall, the results of this study suggest that glucosodiene shows promise as a safe compound for normal cells. However, additional research is needed to fully assess its safety profile and determine its potential therapeutic applications in the context of cancer treatment.

### Acknowledgements

The authors cordially thank the participants for their time and valuable contributions to this research.

# Consent for Publication

Not applicable.

# Data Availability

The datasets used and/or analyzed during the current study are available from the corresponding author upon reasonable request.

# **Competing Interests**

The authors declare that they have no conflicts of interest.

# **Funding**

The authors received no specific funding for this work.

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