CYTOTOXIC EFFECT OF TELOMERASE INHIBITOR MST-312 IN LUNG CANCER CELLS

Villarinho, N.J.[1], Longo, G.M.C.[1], Vasconcelos, F.C.[1], de Oliveira, D.M.[2], Maia, R.C.[1], Lopes, G.P.F.[1]

[1] Laboratório de Hemato-Oncologia Celular e Molecular, PHOM, INCA, RJ, Brazil.

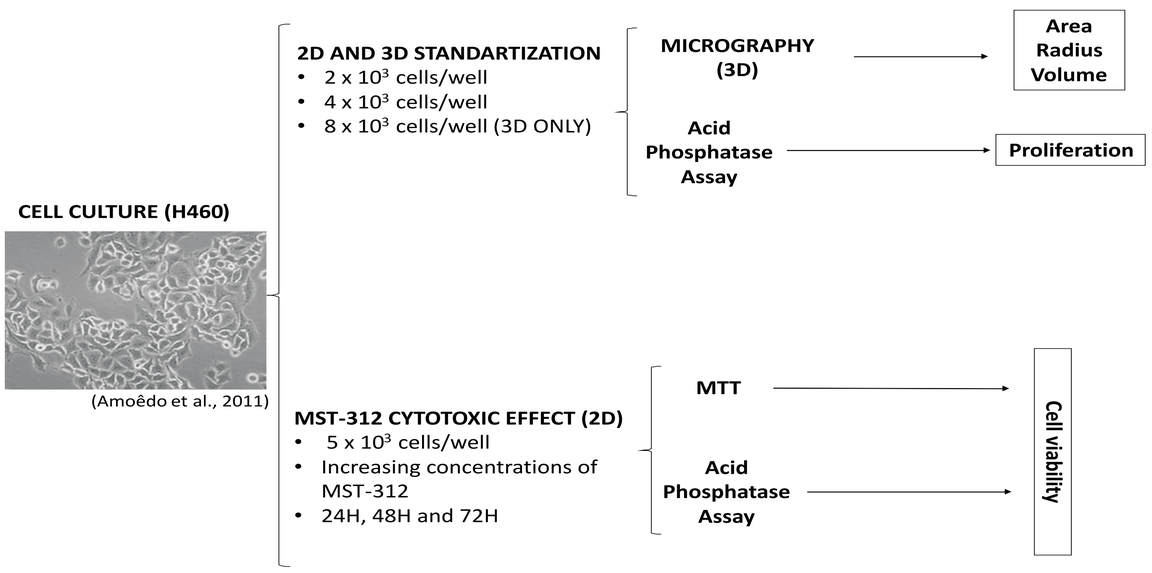
[2] Laboratório de Patologia Molecular, Departamento de Patologia, UnB, DF, Brazil.

INTRODUCTION

In Brazil, lung cancer is the second more incident type of cancer in men and fourth in women in the last year. Cancer cell immortalization by telomeres elongating is one important step in tumorigenesis. A relevant treatment strategy consists in the use of telomerase inhibitors. However, a long-term use of these inhibitors may result in acquired resistance. This resistance is characterized by distinctive mechanisms such as alternative lengthening of telomere (ALT) and the multiple drug resistance (MDR). MDR is related with the expression the *ATP-Binding Cassete* transporters, being overexpressed in Cancer Stem Cells (CSCs). CSCs are a subpopulation of cells that exhibit specific biomarkers and are responsible for tumor recurrence. They also have the ability to form spheroids *in vitro*.

OBJECTIVE

The aim of this research is to develop 2D and 3D models to observe the occurrence cellular immortalization by ALT in lung cancer cells. We intend to evaluate the proliferation profile, CSCs markers and the expression of MDR phenotype during a long-term treatment with MST-312.



RESULTS

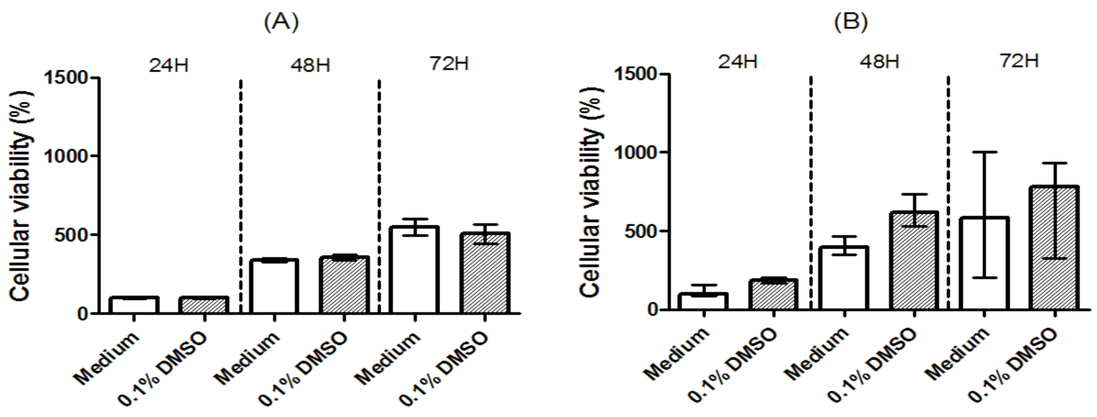


FIGURE 1: H460 cell growth assay seeding (A) 2000 cells and (B) 4000 cells comparing medium supplemented with 10% fetal bovine serum (Control) and vehicle of MST-312 (0.1% DMSO) conditions during 24, 48 and 72h. No significant difference was observed between both conditions. Bars represent the median value and its range representative of eight replicates in an independent experiment.

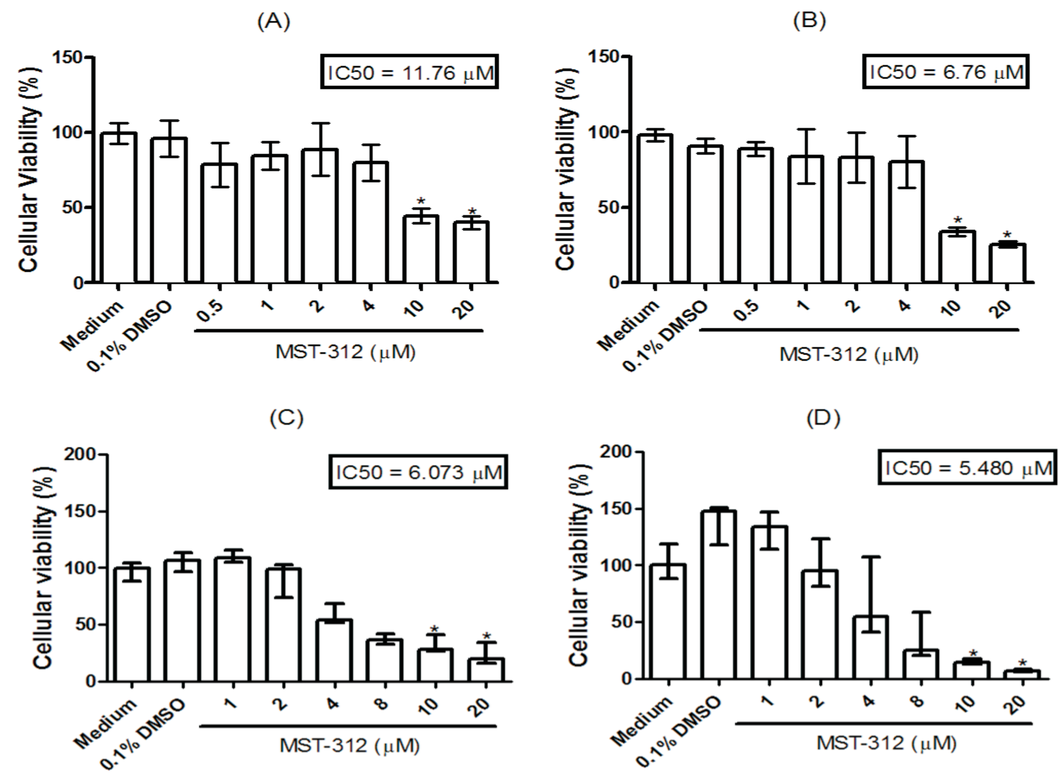


FIGURE 2: Cellular viability assays using H460 and increasing concentrations of the Telomerase Inhibitor MST-312. (A) MTT assay after 48h and (B) 72h of incubation. (C) APH assay after 48h and (D) 72h of incubation. Bars represent the median value and its range representative of eight replicas in three independent experiments.

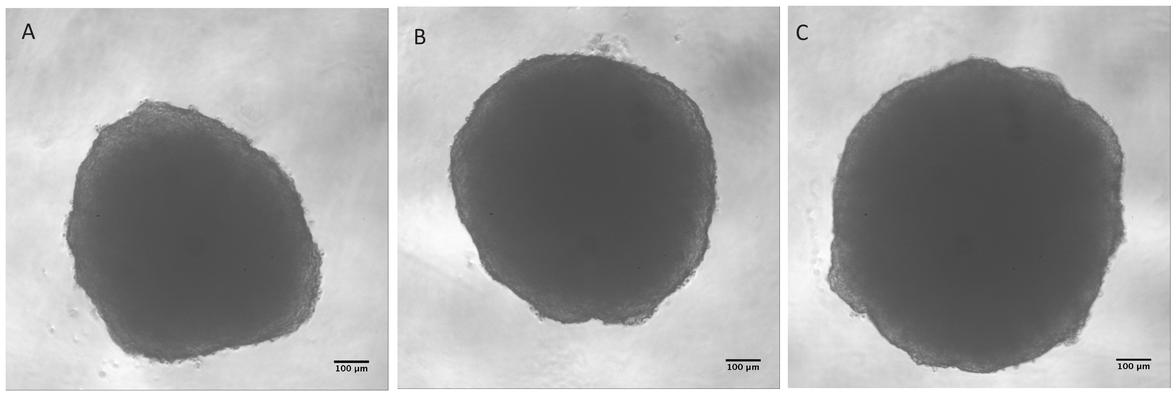


FIGURE 3: Spheroid standardization assay using H460 after 96 hours seeding (A) 2000 cells (B) 4000 cells and (C) 8000 cells.

TABLE 1: Spheroids dimensions.

Cells/ Well	Area (μm^2)	Radius (μm)	Volume (μm^3)
2000	735.337	484	474,344,297.52
4000	830.747	514	569,595,944.56
8000	1,053.477	579	813,395,257.27

CONCLUSION

Before the long-term treatment with MST-312, it was important to determine the specific IC50 and, then, stablish the subtoxic concentration. Starting with the 2D model, the IC50 and subtoxic concentration obtained for H460 was 6.073µM and 2µM, respectively. This data also shows the potential of the chosen drug to decrease the cell viability and supports its potential as a chemotherapic agent. In the next few months, we expect to achieve data concerning the 3D model, start the long-term treatment with both models and its monthly cellular and molecular evaluations concerning CSCs and MDR phenotypes.

KEYWORDS: Lung cancer, Telomerase Inhibitor, Cytotoxicity

FINANCIAL SUPPORT: Ministério da Saúde/INCA and FAPDF

Projeto Gráfico: Setor de Edição e Informação Técnico-Científica / INCA