

# Effect of Plasma Activated Medium on human Head & Neck cancerous Tumor Spheroids.

J.Chauvin<sup>1,2</sup>, F.Judée<sup>1</sup>, N.Merbahi<sup>1</sup>, P.Vicendo<sup>2</sup>

<sup>1</sup> Université de Toulouse - LAPLACE, UPS, Toulouse, France

<sup>2</sup> Université de Toulouse - IMRCP, CNRS, Toulouse, France

This work investigates the effect of Plasma Activated Medium (PAM) on human head and neck cancerous cells using FaDu multicellular tumour spheroids (MCTS). Results indicate that PAM induces cell detachment as soon as the first day post PAM treatment and in a PAM time-dependant manner. The presence of hydrogen peroxide in PAM has been shown to be responsible for this cell detachment. However, a rapid regrowth of the multicellular tumour spheroids size is observed after PAM treatment probably due to a defense mechanism exhibited by FaDu cells. To counteract this effect, successive treatment were done and growth inhibition obtained.

## 1. Introduction

Chemotherapy and radiotherapy have a low rate of success against Head and Neck Cancer due to a high level of resistance [1].

For over a decade, cold atmospheric plasma has been studied in the biomedical field. Its high reactivity allow the creation of RONS [2] that are cytotoxic on various cancer cell lines [3-4] and may induce cell death via apoptosis [4].

Recently, we reported that PAM induced cell death mainly by the involvement of hydrogen peroxide [3].

This work studies the effect of PAM on FaDu MultiCellular Tumor Spheroids (MCTS) and the involvement of hydrogen peroxide in PAM treatment.

## 2. Results

The first effect observed is a volume loss at day one post-treatment associated with a cell detachment. This effect may be attributed to  $H_2O_2$ . The second effect is a rapid re-growth the following days attributed to the auto-organization of MCTS and FaDu defence mechanism (Fig. 1).

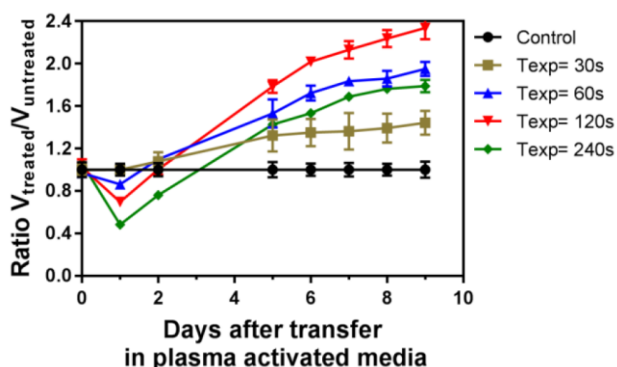


Fig 1. Relative growth of spheroid after PAM treatment for several exposure times.

This increase of spheroids growth may be attributed both to loss of MCTS auto-organization and to FaDu defense mechanisms induced by an external attack.

After 4 successive treatments (Fig. 2), FaDu MCTS were successfully disrupted. However, after each treatment the previously exhibited effects: cell detachment and proliferation were observed until the forth one.

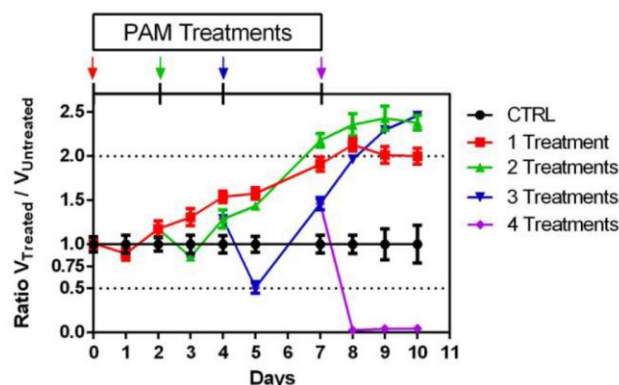


Fig 2. Successive treatment of FaDu spheroids

## 3. References

- [1] Mutschelknaus L, Peters C, Winkler K, Yentrapalli R, Heider T, Atkinson MJ, Exosomes Derived from Squamous Head and Neck Cancer Promote Cell Survival after Ionizing Radiation, *PLoS ONE*, **2016** 11[3]: e0152213
- [2] Fridman, A; Plasma Chemistry, *Cambridge: Cambridge University Press*, **2008**, 1017 p
- [3] Judée, F.; Fongia, C.; Ducommun, B.; Yousfi, M.; Lobjois, V.; Merbahi, N. Short and Long Time Effects of Low Temperature Plasma Activated Media on 3D Multicellular Tumor Spheroids, *Sci Reports*, **2016**, 6, 21421.
- [4] Utsumi, F.; Kajiyama, H.; Nakamura, K.; Tanaka, H.; Hori, M.; Kikkawa, F. Selective Cytotoxicity of Indirect Nonequilibrium Atmospheric Pressure Plasma against Ovarian Clear-Cell Carcinoma, *Springerplus*, **2014**, 3, 398. DOI: 10.1186/2193-1801-3-398