

Introduction

The exosome, a small extracellular vesicle, is being studied as a means of intercellular communication. These microvesicles aid in the transport of proteins, microRNA, DNA fragments, and more between cells (Figure 1). Studies have shown that when they are released into the microenvironment, they induce changes in neighboring cells that promote tumor formation and inhibit anti-tumor immune responses, resulting in cancer cell migration, no immune detection, and established metastatic growth (2).

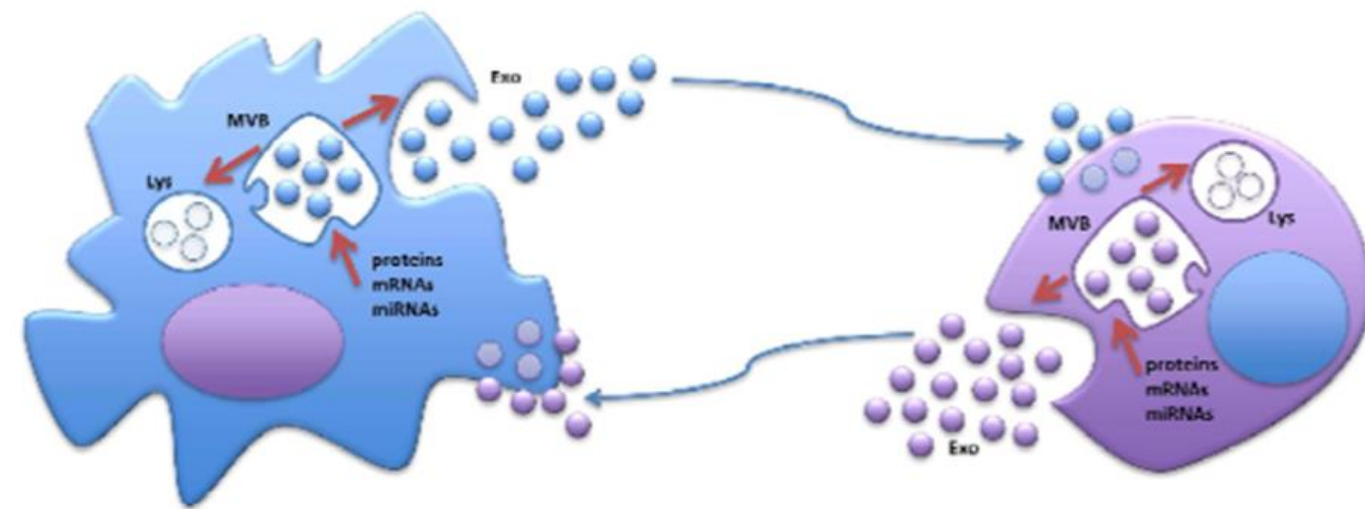


Figure 1 Exosome Crosstalk (1)

This study examines the role of exosomes in the proliferation and growth of HPV negative cervical cancer. Cervical cancer often arises after infection with Human Papilloma Virus (HPV). However, there are instances where cervical cancer has arisen spontaneously without the presence of HPV (3), and a better understanding of the molecular mechanisms of HPV negative cell growth is needed so that better treatment options can be established.

References

1. Neviani, P., & Fabbri, M. (2015). Exosomal microRNAs in the Tumor Microenvironment. *Frontiers in Medicine*, 2, 47.
2. Tickner, Jacob A., Aaron J. Urquhart, Sally-Anne Stephenson, Derek J. Richard, and Kenneth J. O'Byrne. Functions and Therapeutic Roles of Exosomes in Cancer. *Oncol. Frontiers in Oncology*. 2014; 127: 1-8.
3. Walboomers, Jan M. M, Marcel V. Jacobs, Michelle Manos, et, al. Human Papillomavirus is a Necessary Cause of Invasive Cervical Cancer Worldwide. *J. Pathol.* 1999; 189: 12-19.

Results

Determination of optimal plating density of WI-38

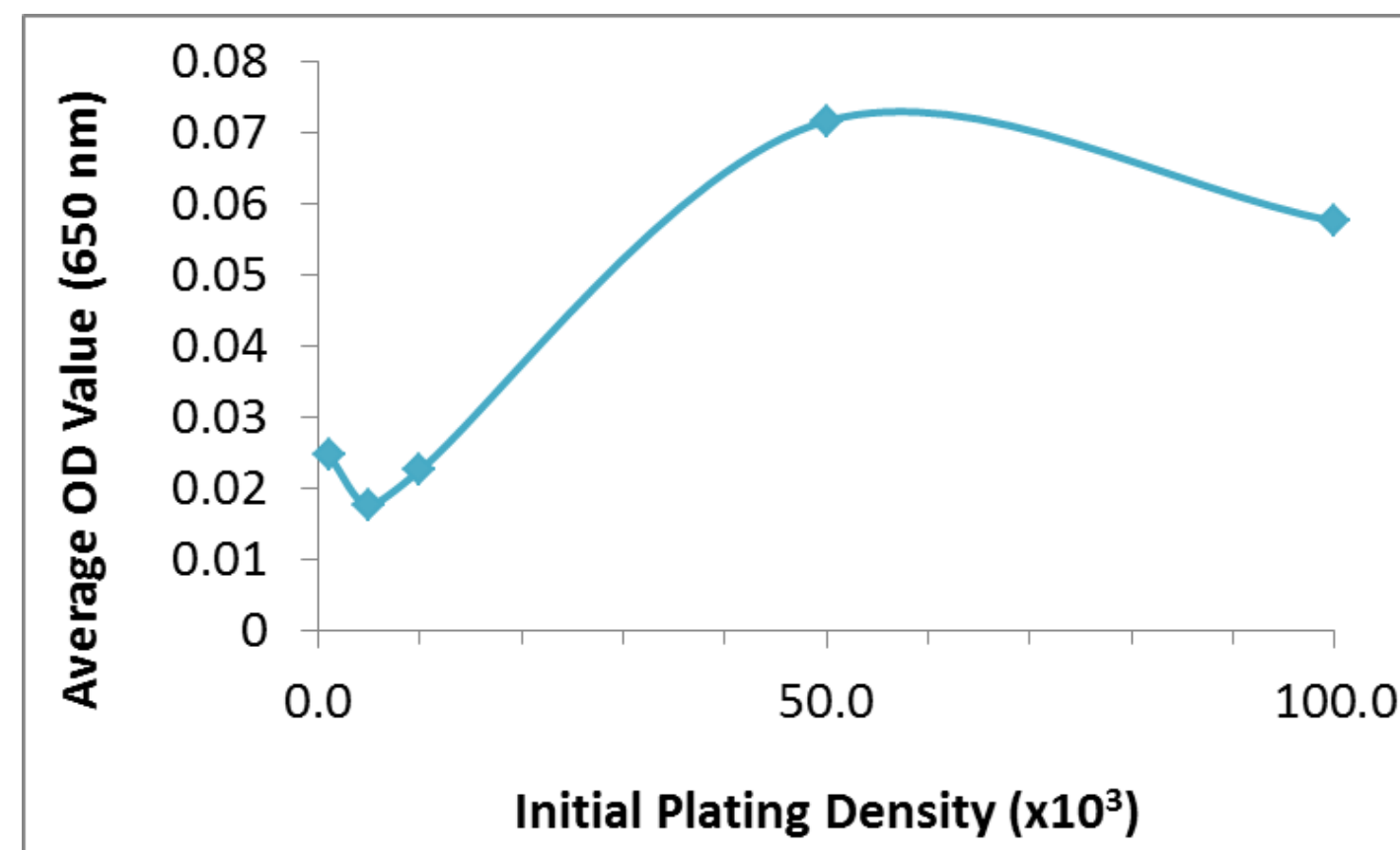


Figure 2 WI-38 cell viability was measured 24 hours after plating cells at the indicated cell density (n=2) by MTT viability assay.

Exosome treatment inhibits cell proliferation and causes an increase in cell size

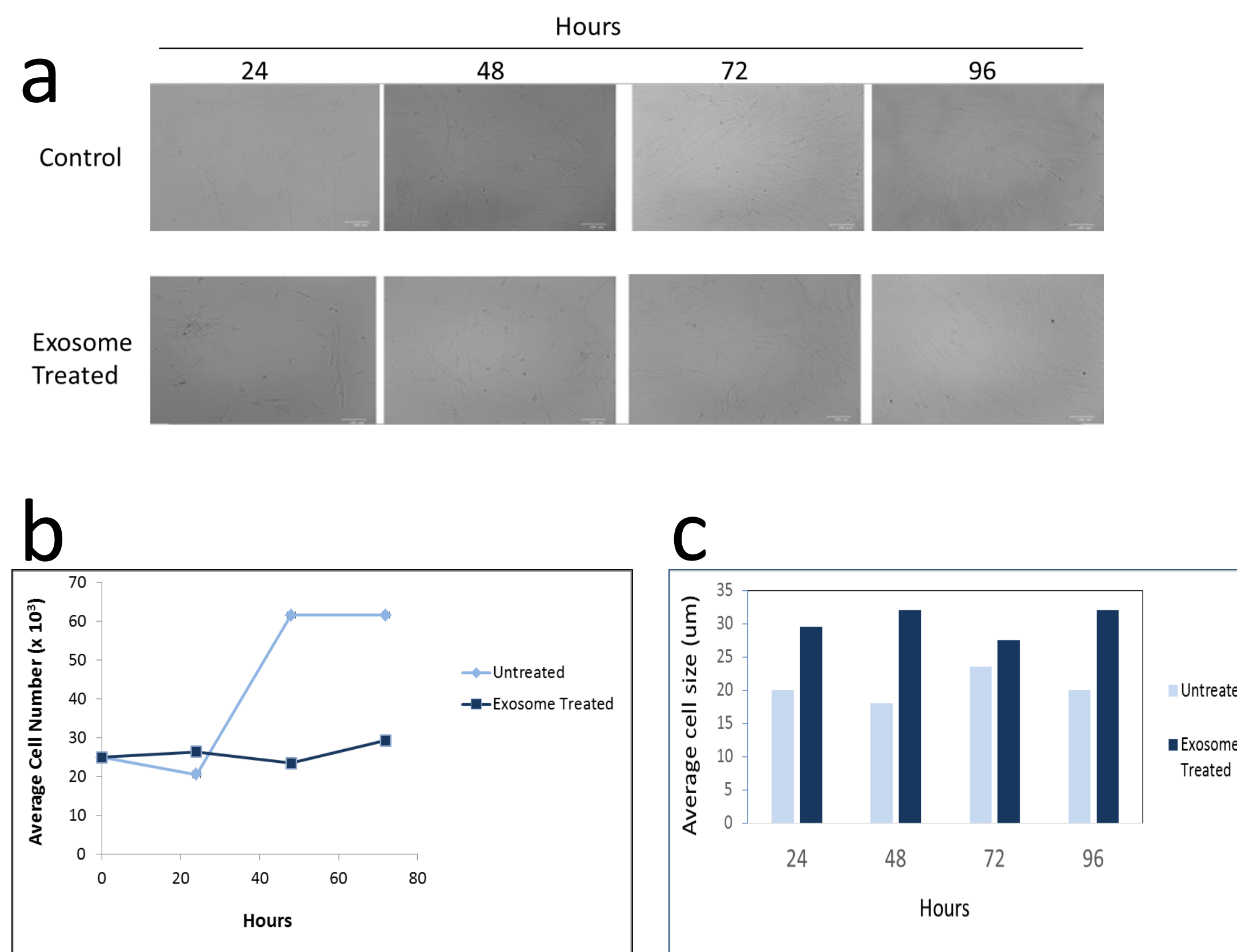


Figure 3 WI-38 cells were left untreated or treated with DoTC-4510 exosomes. At the indicated time points, (a) photographs were taken using the ZOE Cell Imager (b) cells were counted (n=2) and (c) cell size was measured using the Countess Automated Cell Counter (n=1).

Methods

Exosome Isolation

- Conditioned media (CM) was collected from DOTC 4510 HPV negative cervical cancer cell line
- Exosomes were isolated by filtering CM through 100 kda MWCO Amicon filters followed by pelleting at 10,000 x g
- Purification of exosomes was verified by western blot for the exosomal marker CD63 (data not shown)

Exosome Treatments and Proliferation Assay

- Wi-38 normal lung epithelial cells, were left untreated or treated with DoTC-4510 exosomes
- Viability was measured using the MTT assay
- Proliferation was measured by counting cells every day for five days
- Cell size was measured using the Countess Automated cell counter

Discussion

- Exosomes were successfully isolated from DoTC-4510 cells
- Exosomes did not cause a change in the viability of normal epithelial cells
- Exosomes treatment did cause a decrease in proliferation and an increase in cell size. Normal epithelial cells treated with exosomes were larger than the untreated cells, indicated that exosomes may affect cell cycle progression.

Further studies

- Repeat the results to determine statistical significance
- Examine cell cycle progression after exosome treatment

Acknowledgements

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