

VIEWPOINT

Stromal-cell and cancer-cell exosomes leading the metastatic exodus for the promised niche

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Abstract

Exosomes are thought to play an important role in metastasis. Luga and colleagues have described the production of exosomes by stromal cells such as cancer-associated fibroblasts that are taken up by breast cancer cells and are then loaded with Wnt 11, which is associated with stimulation of the invasiveness and metastasis of the breast cancer cells. Previous studies have shown that exosomes produced by breast cancer cells are taken up by stromal fibroblasts and other stromal cells, suggesting that exosomes are agents of cross-talk between cancer and stromal cells to stimulate metastasis. Imaging of exosomes by labeling with fluorescent proteins will enlighten the process by which exosomes enhance metastasis, including premetastatic niche formation.

Exosomes are small (30 to 100 nm) membrane vesicles that originate from the endosomal membrane compartment [1-3]. Exosomes contain mRNAs, miRNAs and proteins, and possibly other components as well [1,4,5]. Cancer cells release exosomes into the tumor microenvironment and peripheral blood [1,6].

Do stromal cells in the tumor microenvironment also produce exosomes that stimulate cancer metastasis? We previously showed that cancer cell-associated stromal cells are necessary for metastasis to occur [7]. Luga and colleagues now report in a recent issue of *Cell* that stromal cells indeed produce exosomes and that fibroblast-secreted exosomes promote breast cancer cell (BCC) motility and metastasis by mobilizing autocrine Wnt11-induced planar cell polarity in the cancer cells [8]. Wnt11 is tethered to the fibroblast-derived exosomes within BCCs. Exosome stimulation of BCC invasion and metastasis was shown to be dependent on Wnt11

produced in the BCCs. Cancer-associated fibroblast (CAF)-derived exosomes were internalized by BCCs and then loaded with Wnt11 in orthotopic mouse models of breast cancer. Luga and colleagues report that exosomes secreted from human breast CAFs stimulate BCC protrusive activity, motility, and metastasis. These properties are dependent on the exosome tetraspanin, Cd81, which is critical for exosome-stimulated BCC metastasis [8]. These are intriguing findings on how stromal cells promote metastasis via exosomes.

Previous suggestions indicated that recruited bone marrow progenitor cells generated a premetastatic niche to which the cancer cells metastasize [9-11]. Secreted factors in the bone marrow may be important to mobilize cells to form the premetastatic niche. Exosomes, derived from cancer cells, have recently been shown to have an important role in premetastatic niche formation [9-11]. However, Luga and colleagues do not suggest how stromal cell-derived exosomes play a role in the formation of a premetastatic niche [8].

Investigating premetastatic niche formation requires the possibility for dynamic imaging of exosomes *in vivo*. To image the fate of cancer cell-derived exosomes in orthotopic nude mouse models of breast cancer, we used GFP-tagged CD63, which is a general marker of exosomes [12]. BCCs were imaged to transfer their own exosomes to other cancer cells and normal lung tissue cells in culture. In orthotopic nude mouse models, BCCs secreted exosomes into the tumor microenvironment. Tumor-derived exosomes were incorporated into tumor-associated cells at a metastatic site, including CAFs (Figure 1), and in the circulation. These results suggest that tumor-derived exosomes may contribute to forming a niche to promote tumor growth and metastasis. Our results demonstrate the usefulness of GFP imaging to investigate the role of exosomes in cancer metastasis [12-15].

Both cancer cell-derived or stromal cell-derived exosomes are thus able to alter the tumor environment and may participate in forming a distant metastatic niche to promote metastasis. Dynamic imaging of exosomes derived from cancer or stromal cells in metastatic models may hence help us to understand the mechanism of

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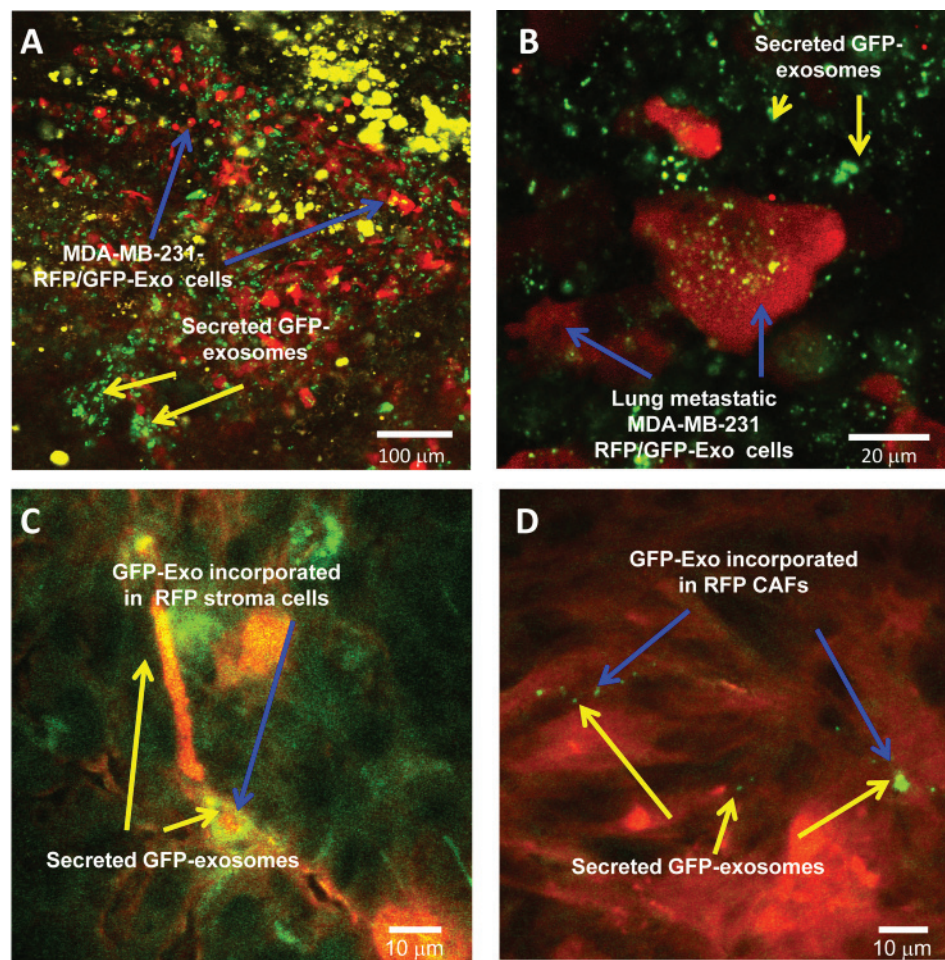


Figure 1. Cancer cells secrete exosomes into the tumor microenvironment in human MDA-MB-231 breast cancer orthotopic mouse models. The red fluorescent protein (RFP)-expressing MDA-MB-231-cells-produced exosomes which were labeled with a CD63-GFP fusion protein (MDA-MB-231-RFP/GFP-Exo). **(A)** MDA-MB-231-RFP/GFP-Exo cells secreted GFP exosomes in the primary tumor tissue. Blue arrows, MDA-MB-231-RFP/GFP-Exo cells; yellow arrows, secreted GFP exosomes. Scale bar = 100 μm. **(B)** MDA-MB-231-RFP/GFP-Exo cells secreted GFP exosomes at the lung colonization site. Yellow arrows, secreted GFP exosomes; blue arrows, lung metastatic MDA-MB-231-RFP/GFP-Exo cells. Scale bar = 20 μm. **(C)** Blue arrows, GFP-Exo incorporated in RFP stroma cells; yellow arrows, secreted GFP exosomes. Scale bar = 10 μm. **(D)** Blue arrows, GFP-Exo incorporated in RFP cancer-associated fibroblasts (CAFs); yellow arrows, secreted GFP exosomes. Scale bar = 10 μm [12].

cancer metastasis. Imaging of exosomes may also be useful to predict the location of future metastasis in real time.

This viewpoint demonstrates the importance of exosome cross-talk between cancer cells and stromal cells. Luga and colleagues demonstrate the production of exosomes by stromal cells such as CAFs that are taken up by BCCs, which in turn promote their invasive and metastatic activity [8]. Suetsugu and colleagues demonstrate production of exosomes by BCCs that are taken up by CAFs [10], the reciprocal of what was observed by Luga and colleagues [8]. With the use of fluorescent-protein *in vivo* imaging [13-15], further understanding of exosomes and their relationship to metastasis, including niche formation, will surely be enlightened.

Abbreviations

BCC, breast cancer cell; CAF, cancer-associated fibroblast; GFP, green fluorescent protein; miRNA, microRNA.

Competing interests

The author declares that he has no competing interests.

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