Chemoresistance to paclitaxel induces EMT in different types of ovarian carcinoma tumors

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Introduction

Ovarian cancer — 140.000 deaths/year worldwide — 45% of survival

Lack of measurable early symptoms → Advanced stage at diagnosis

Treatment → Removal/debulking surgery

Treatment → Paclitaxel + cisplatin chemotherapy → Chemoresistance → EMT

Epithelial-mesenchymal transition (EMT) → Metastasis

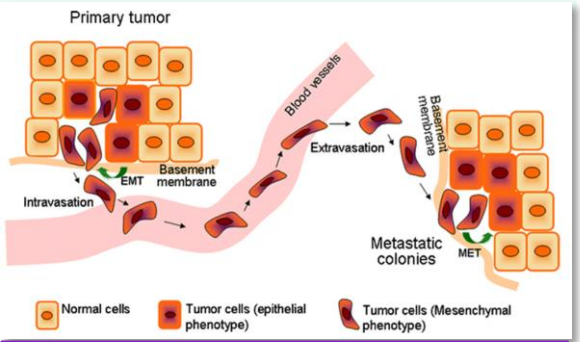

Fig. 1 Epithelial-mesenchymal transition process in cancer: from a primary tumor epithelial cell to a motile mesenchymal cell. Meng F et al.

Hypothesis and Objectives

The chemoresistance to placlitaxel promotes the epithelial-mesenchymal transition in the four main epithelial ovarian tumor types by the upregulation of the transcription factors that repress E-cadherin.

Chemoresistance to paclitaxel → ↑ Snail, Slug, Twist1, Zeb1 and Zeb2 → ↓ E-cadherin → EMT

- Analyze paclitaxel resistant ovarian carcinoma cells: phenotype, migration, proliferation, gene expression.
- Determine if there is a relation between the activation of the transcription factors and the acquisition of paclitaxel chemoresistance.
- Compare the results of the different tumor types.

Material and methods

- Human cell lines
  - Serous ovarian adenocarcinoma from ascites (OV17R)
  - Mucinous ovarian carcinoma (COV644)
  - Endometrioid ovarian carcinoma (COV362)
  - Clear cell ovarian carcinoma(ES-2)
- Establishment of paclitaxel resistance and chemosensitivity assay: IC50
- Proliferation: MTT assay
- Anchorage-independent growth: Soft agar assay
- Invasion: Boyden chamber assay
- Migration: Wound-healing assay
- Immunofluorescence
- Western Blot
- Statistical analysis

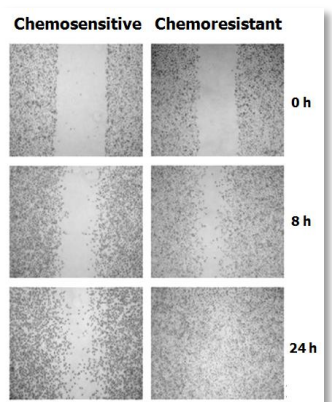

Fig. 5 Wound healing assay. Modified from Yang YC et al.

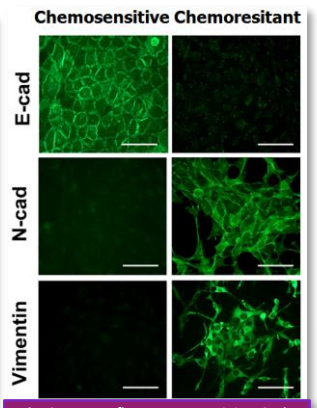

Fig. 6 Immunofluorescence staining. Scale bars, 100 µm. Modified from Evdokimova et al.

Expected results

	Chemosensitive cells	Chemoresistant cells
Chemosensitivity	↓ IC50	↑ IC50
Proliferation	↑ Cell number	↓ Cell number
Anchorage-independent growth	↓ Colonies	↑ Colonies
Invasion	↓ Cell invasion	↑ Cell invasion
Migration	↓ Cell migration	↑ Cell migration
Immunofluorescence	↑E-cadherin, ↓N-cadherin, ↓Vimentin	↓E-cadherin, ↑N-cadherin, ↑Vimentin
Western Blot	↓ Gene expression	↑ Gene expression

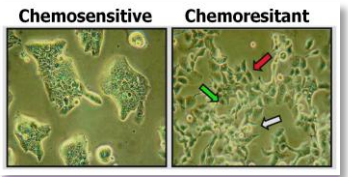

Fig. 2 Morphologic changes. Spindle-cell shaped cells with loss of polarity (red), intercellular separation (green), and pseudopodia (white). x20 magnifications. Modified from Yang AD et al.

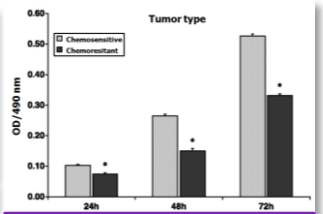

Fig. 3 Proliferation assay. Modified from Yang AD et al.

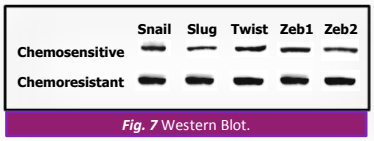

Fig. 7 Western Blot.

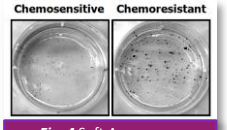

Fig. 4 Soft Agar assay. Modified from Dallas et al.

Benefits

A better understanding of the mechanisms that underlie the chemoresistance by which tumor cells survive treatment could lead to the identification of novel therapeutic targets and development of an appropriate therapy for certain cancers, like ovarian carcinoma, for which the early detection is still a barrier.

References

1. Meng F and Wu G (2012). The rejuvenated scenario of epithelial-mesenchymal transition (EMT) and cancer metastasis. *Can Met Rev* 31(3-4):455-467.
2. Evdokimova V, Tognon C, Ng T, et al. (2009). Translational Activation of Snail1 and Other Developmentally Regulated Transcription Factors by YB-1 Promotes an Epithelial-Mesenchymal Transition. *Cancer Cell* 15(5):402-415.
3. Yang AD, Fan F, Camp ER et al. (2006). Chronic oxaliplatin resistance induces epithelial-to-mesenchymal transition in colorectal cancer cell lines. *Clin Cancer Res* 12:4147-4153.
4. Yang YC, Ho TC, Che SL, et al. (2007) Inhibition of cell motility by troglitazone in human ovarian carcinoma cell line. *BMC Cancer* 7:216.
5. Dallas NA, Xia L, Fan F, et al.. (2009) Chemoresistant colorectal cancer cells, the cancer stem cell phenotype, and increased sensitivity to insulin-like growth factor-I receptor inhibition. *Cancer Res* 69:1951-1957.