Chemioterapia tollerabile: un ossimoro superabile con la sinergia?

DiSIT - Dipartimento di Scienze ed Innovazione Tecnologica
Alessandria

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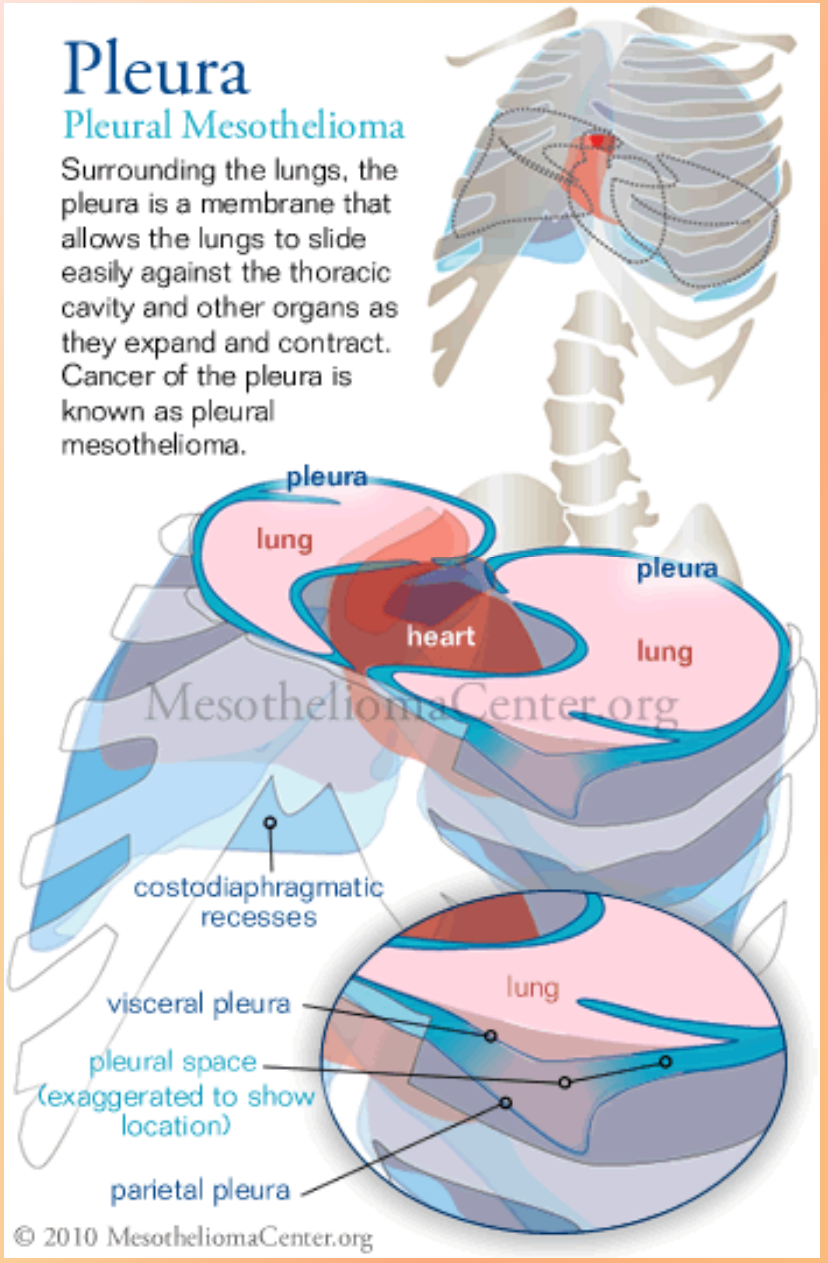

Pleura
Pleural Mesothelioma
Surrounding the lungs, the pleura is a membrane that allows the lungs to slide easily against the thoracic cavity and other organs as they expand and contract. Cancer of the pleura is known as pleural mesothelioma.
pleura
lung
pleura
heart
lung
MesotheliomaCenter.org
costodiaphragmatic recesses
lung
visceral pleura
pleural space (exaggerated to show location)
parietal pleura
© 2010 MesotheliomaCenter.org

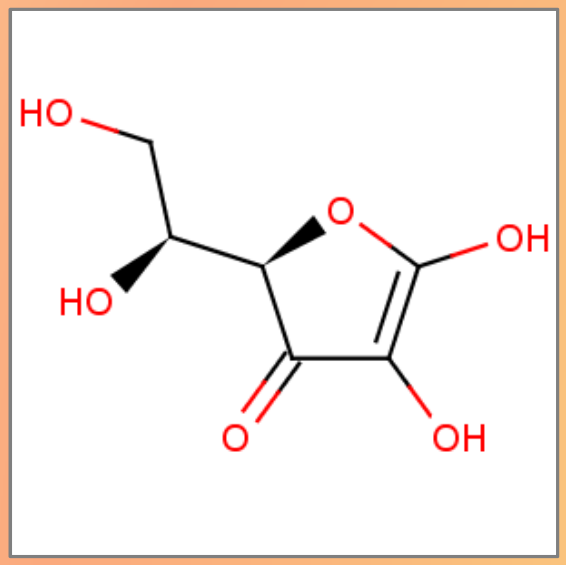

DANGER
ASBESTOS
DANGER
ASBESTOS


CAUTION
ASBESTOS
HAZARD
DO NOT DISTURB
WITHOUT PROPER
TRAINING AND
EQUIPMENT

Sono necessarie nuove strategie!!!

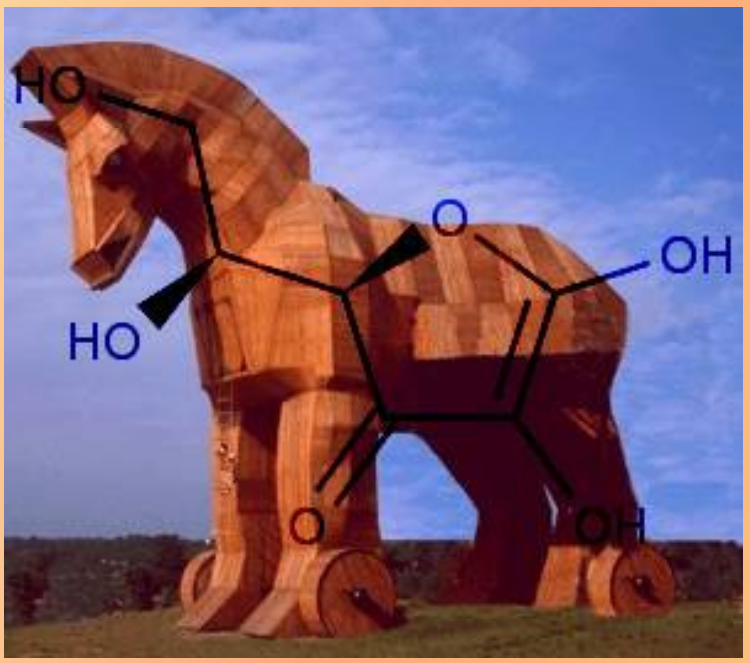
Acido Ascorbico

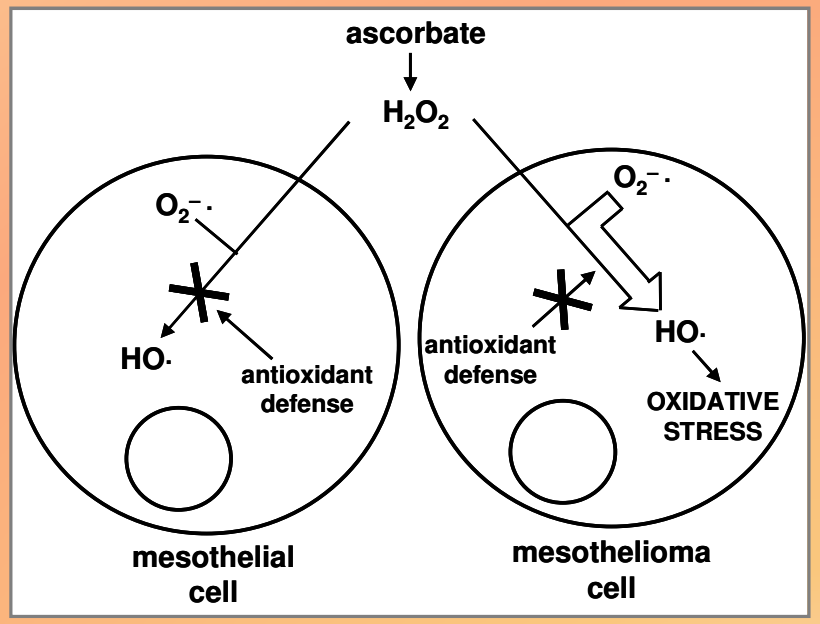
- ☑ L'ascorbato è un nutriente essenziale nella dieta
- ☑ Già 50 anni fa, venne proposto nel trattamento dei tumori.
- ☑ Grazie alla forte azione riducente, la vitamina C è utilizzata in molte reazioni di ossidoriduzione.



Ascorbato

L'ascorbato è più tossico sulle cellule di mesotelioma rispetto a quelle di mesotelio per la produzione extracellulare di H_2O_2

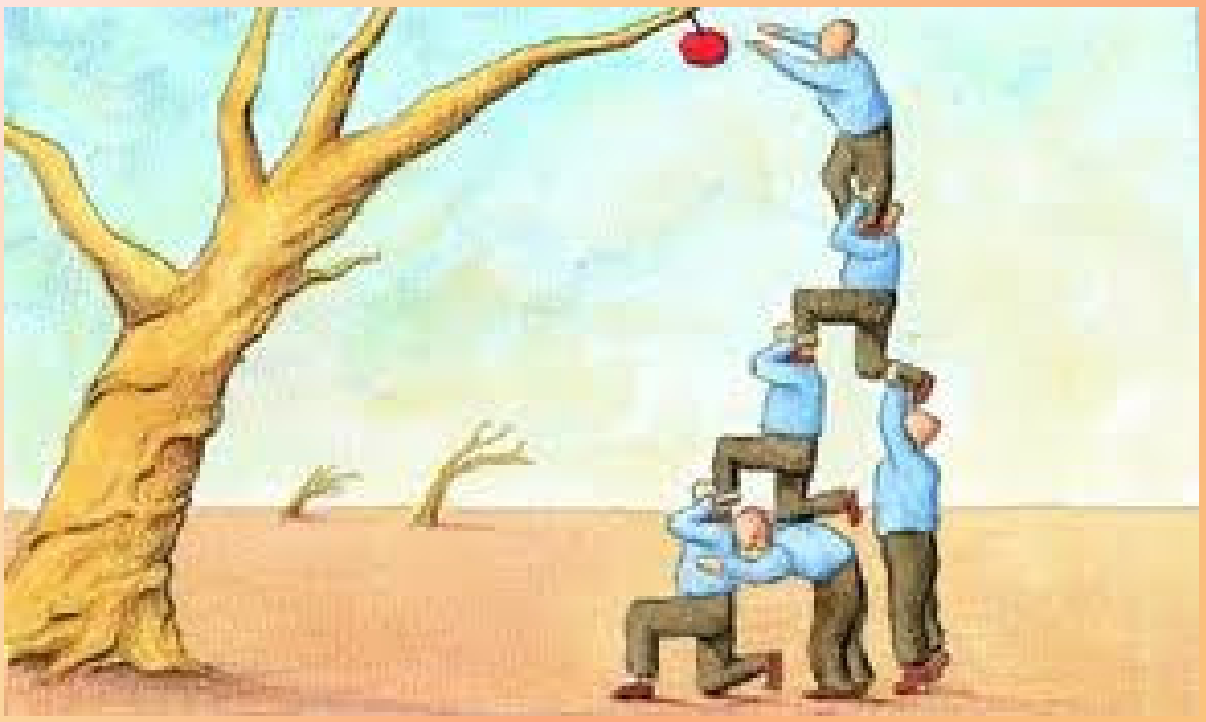




Ranzato *et al*., Am J Respir Cell Mol Biol 2011

L'H_2O_2 prodotto nello spazio extracellulare reagisce con i radicali superossido generati dalle cellule di mesotelioma, causando un importante stress ossidativo intracellulare, in grado di indurre danno alle cellule.

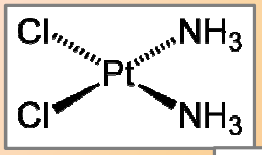
L'ascorbato agisce con un meccanismo a cavallo di Troia

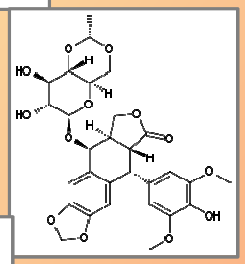


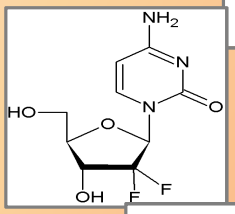
Sinergia

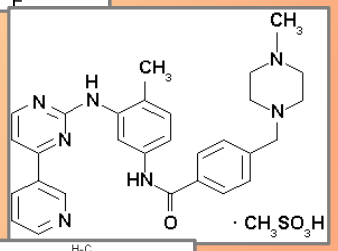
La sinergia (dal greco συνεργός, che significa "lavorare insieme"), è definita come la reazione di due o più agenti che lavorano insieme per produrre un risultato non ottenibile singolarmente

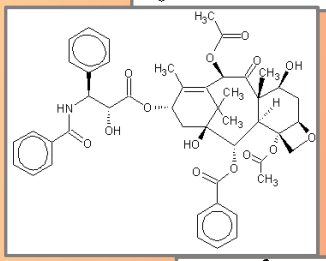
Drug	Treatment length	EC50 µM	Lower CI 95%	Upper CI 95%
Cis-Platin	48 h	11.92	9.79	14.52
Etoposide	24 h	39.41	27.36	56.77
Gemcitabine	48 h	67.99	49.56	93.28
Imatinib	24 h	71.29	56.08	90.61
Paclitaxel	24 h	1.16	0.43	3.13
Taurolidine	24 h	68.94	57.15	83.17
Tyrphostin	24h	45.76	40.11	52.22
Tocopherol	24h	12.55	8.50	18.52
EGCG	24h	103.3	80	133

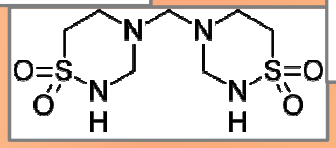


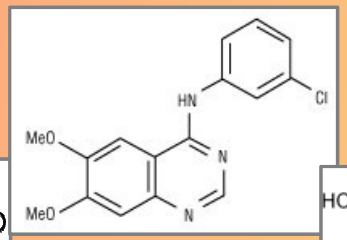


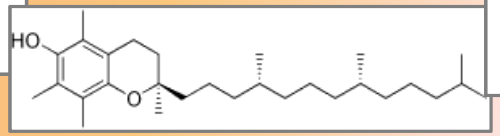


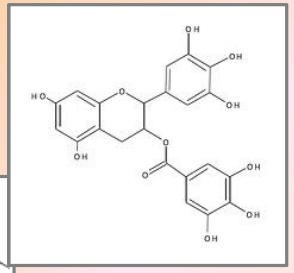








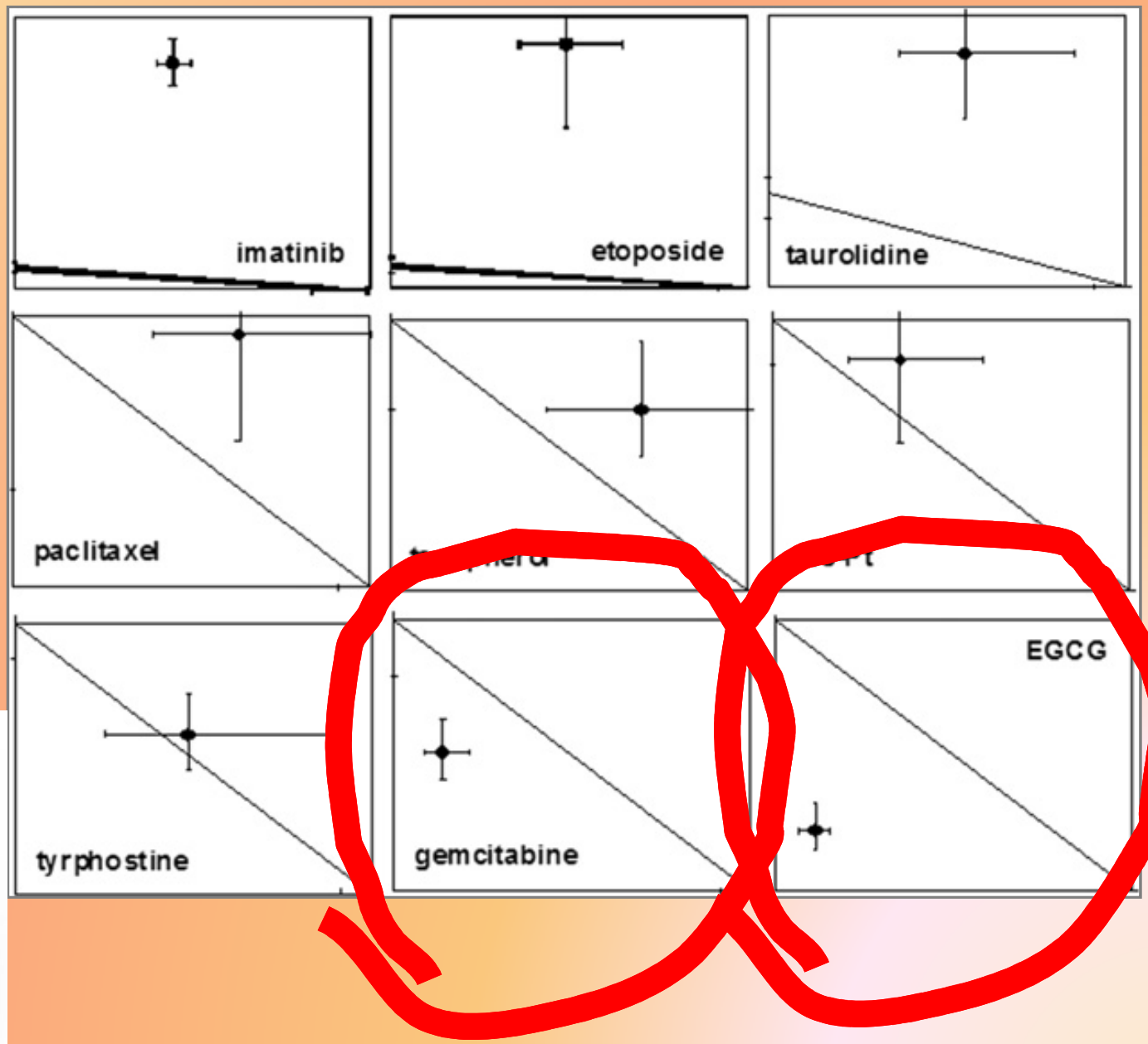




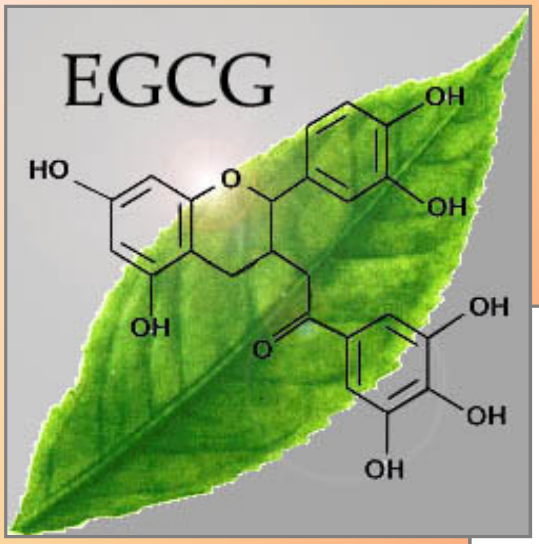
Martinotti *et al.*, Toxicology in Vitro, 2011

Martinotti *et al*., Toxicology in Vitro, 2011

L'analisi di Isobologramma



Epigallocatechina-3-gallato



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Epigallocatechin-3-gallate induces mesothelioma cell death *via* H_2O_2–dependent T-type Ca^{2+} channel opening

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Abstract

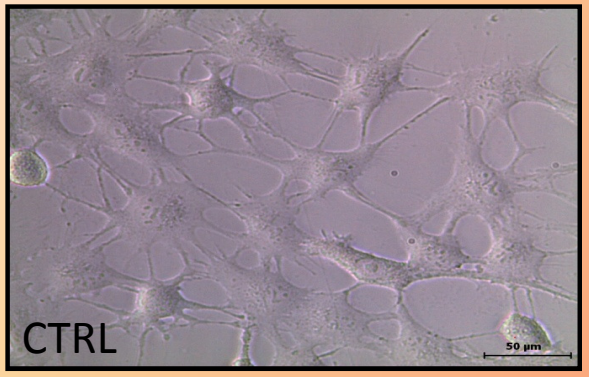
Malignant mesothelioma (MMe) is a highly aggressive, lethal tumour requiring the development of more effective therapies. The green tea polyphenol epigallocathechin-3-gallate (EGCG) inhibits the growth of many types of cancer cells. We found that EGCG is selectively cytotoxic to MMe cells with respect to normal mesothelial cells. MMe cell viability was inhibited by predominant induction of apoptosis at lower doses and necrosis at higher doses. EGCG elicited H_2O_2 release in cell cultures, and exogenous catalase (CAT) abrogated EGCG-induced cytotoxicity, apoptosis and necrosis. Confocal imaging of fluo 3-loaded, EGCG-exposed MMe cells showed significant $[Ca^{2+}]_i$ rise, prevented by CAT, dithiothreitol or the T-type Ca^{2+} channel blockers mibefradil and $NiCl_2$. Cell loading with dihydrorhodamine 123 revealed EGCG-induced ROS production, prevented by CAT, mibefradil or the Ca^{2+} chelator BAPTA-AM. Direct exposure of cells to H_2O_2 produced similar effects on Ca^{2+} and ROS, and these effects were prevented by the same inhibitors. Sensitivity of REN cells to EGCG was correlated with higher expression of $Ca_v3.2$ T-type Ca^{2+} channels in these cells, compared to normal mesothelium. Also, $Ca_v3.2$ siRNA on MMe cells reduced *in vitro* EGCG cytotoxicity and abated apoptosis and necrosis. Intriguingly, $Ca_v3.2$ expression was observed in malignant pleural mesothelioma biopsies from patients, but not in normal pleura. In conclusion, data showed the expression of T-type Ca^{2+} channels in MMe tissue and their role in EGCG selective cytotoxicity to MMe cells, suggesting the possible use of these channels as a novel MMe pharmacological target.

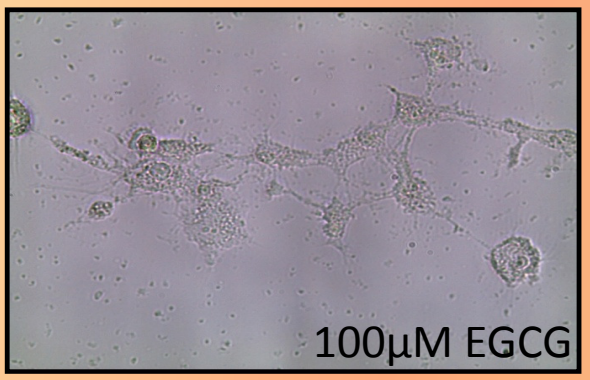
Keywords: anticancer therapy • hydrogen peroxide • reactive oxygen species • malignant mesothelioma • T-type calcium channels

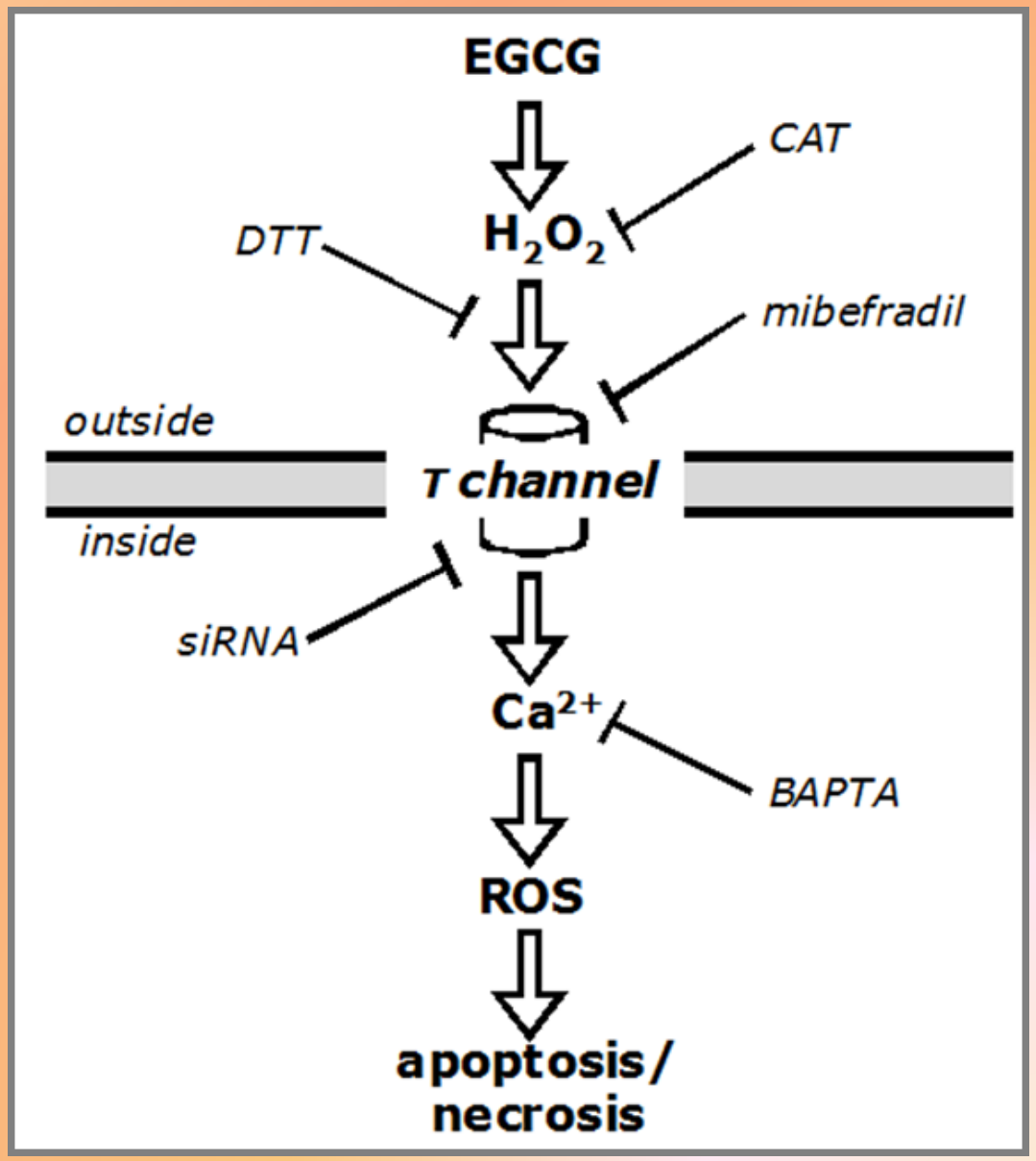
Introduction

Malignant mesothelioma (MMe) is a highly aggressive tumour arising from mesothelial cells of the serosal surfaces of body cavities, mainly

Chemotherapy is the most widely used clinical approach, with various cytotoxic drugs having already been tested [2–4]. The response rate



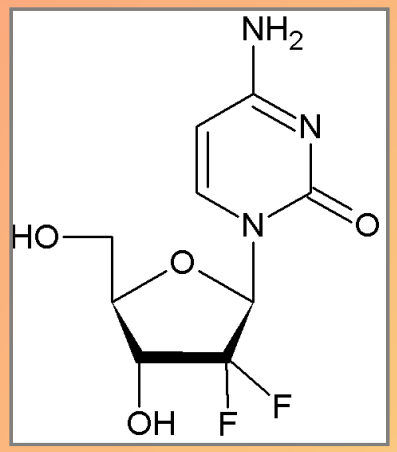




Ranzato, Martinotti *et al.*, JCMM 2012

…i tre attori principali della nostra ricerca

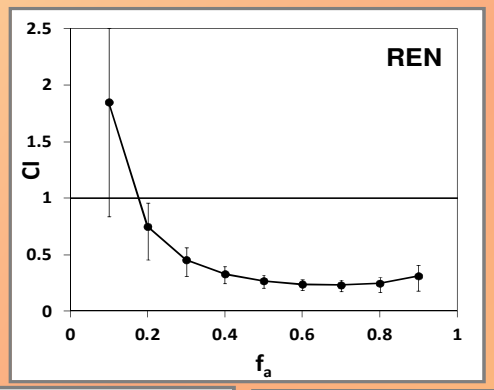
Ascorbato

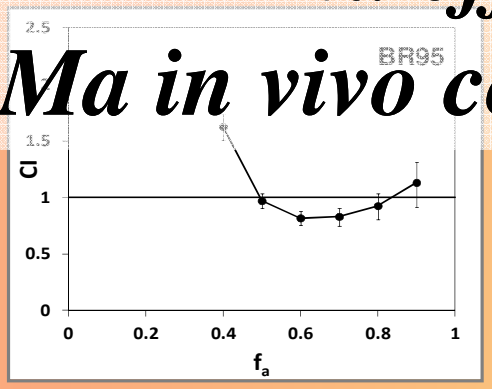


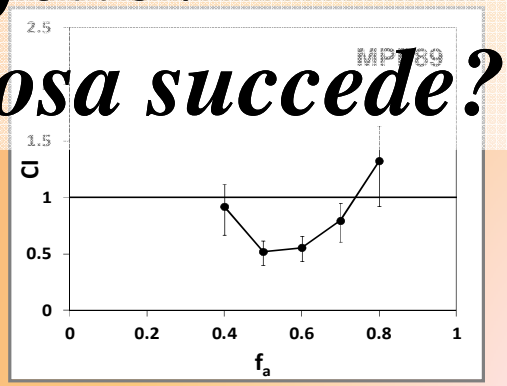
Gemcitabina

EGCG

In vitro drug interaction analysis



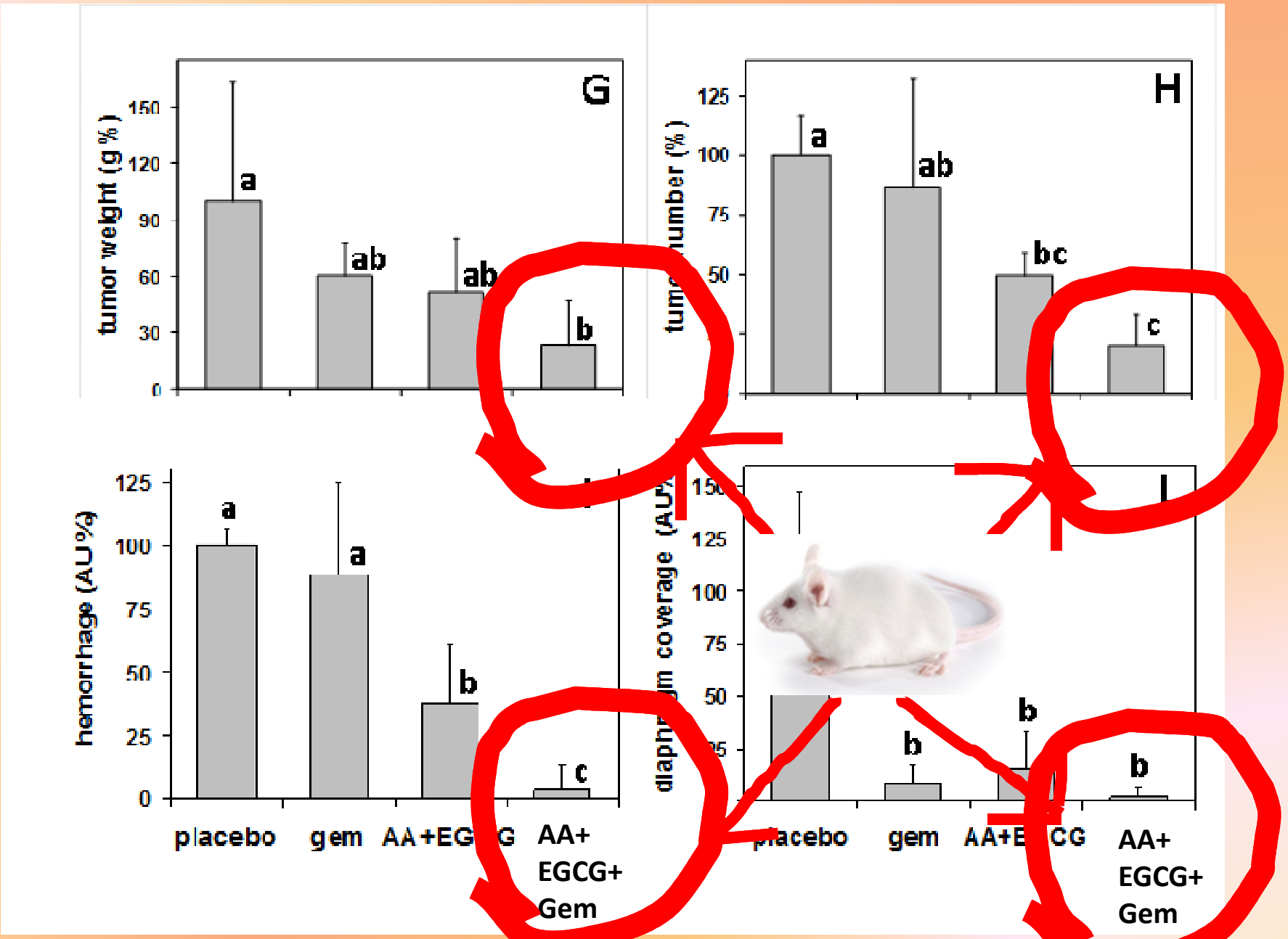




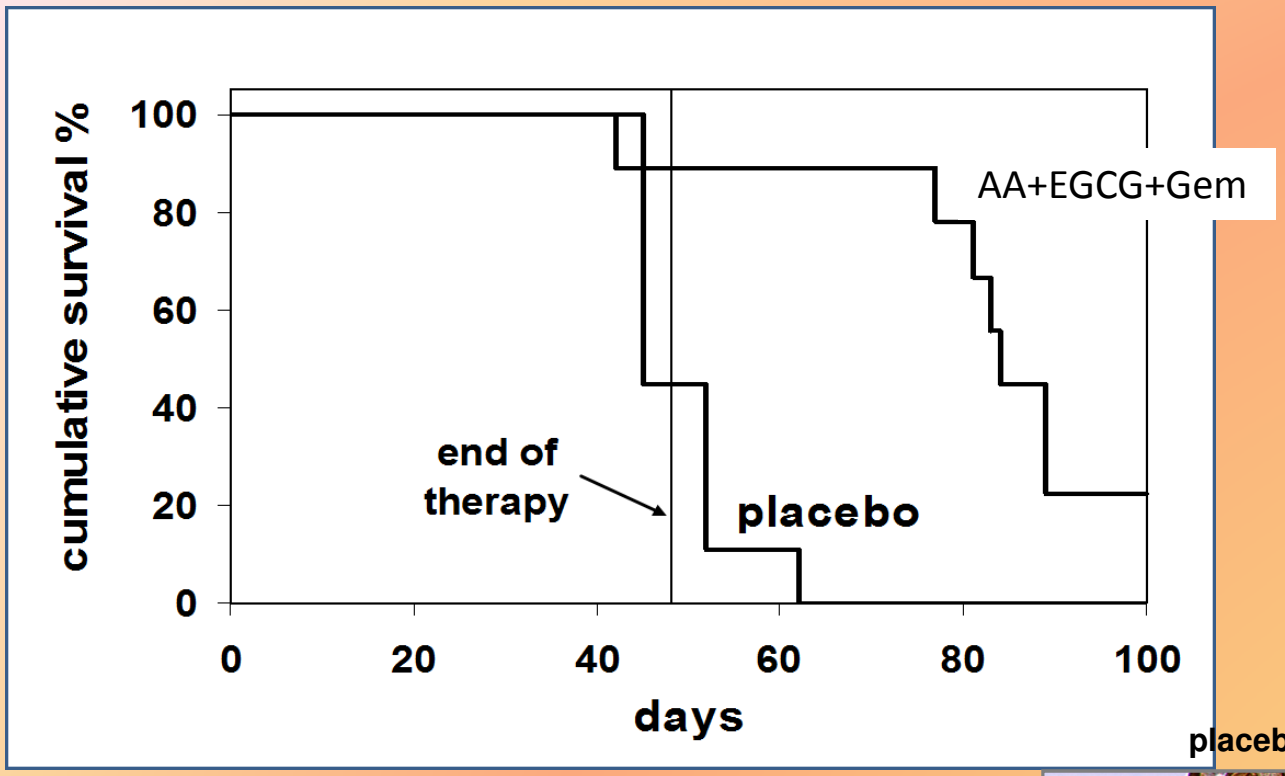
Negli esperimenti in vitro la combinazione dei tre composti ha effetto!

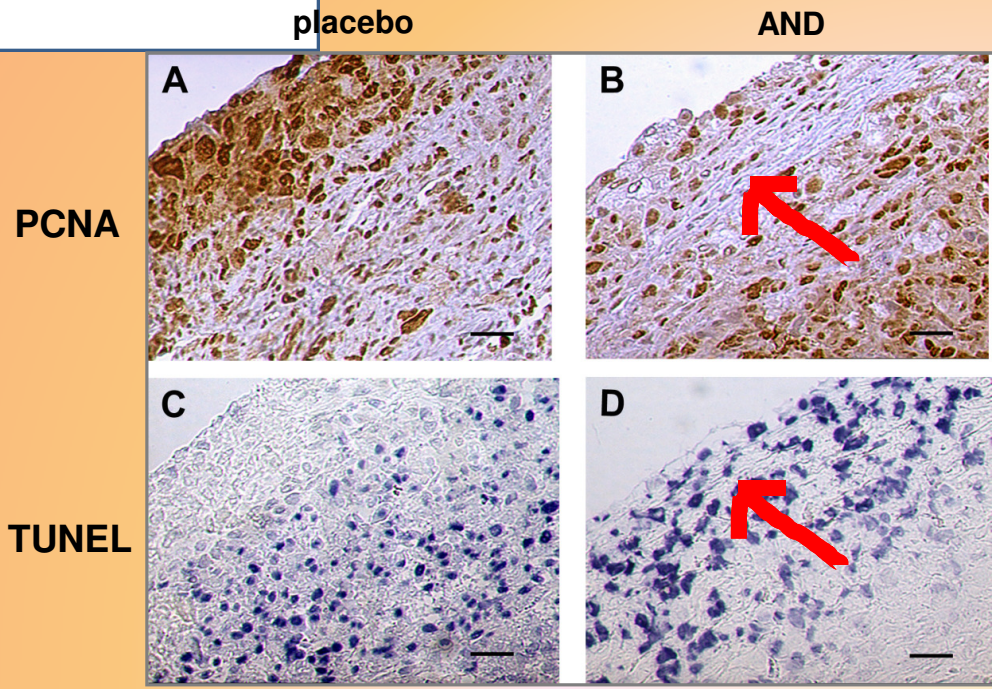
Ma in vivo cosa succede?

Volta, Ranzato *et al.*,
PLoS ONE under review



Volta, Ranzato *et al.*,
PLoS ONE under review

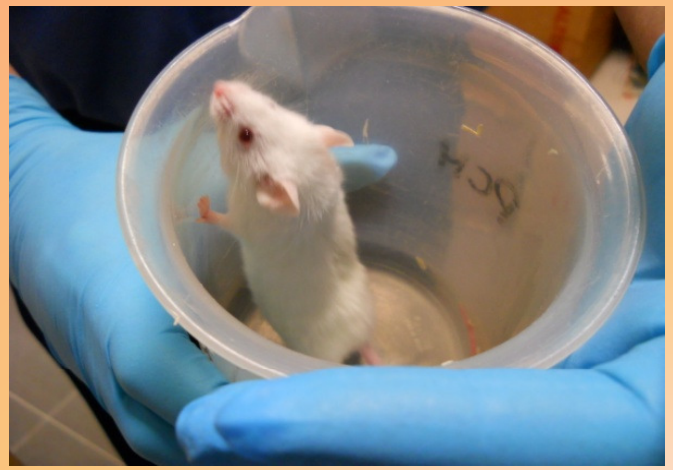

cumulative survival %
100
80
60
40
20
0
0
20
40
60
80
100
days
AA+EGCG+Gem
end of
therapy
placebo

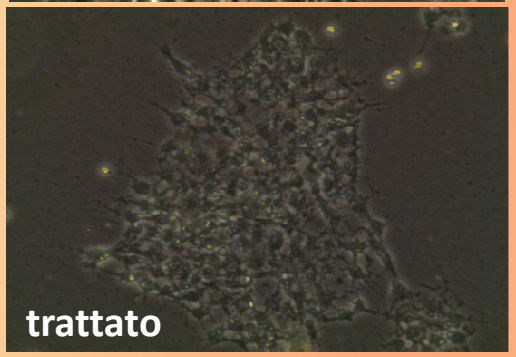

placebo
AND
PCNA
TUNEL
A
B
C
D

Conclusioni

Due composti sono sinergisticamente attivi con l'acido ascorbico nelle analisi *in vitro*: l'**epigallocatechina-3-gallato** e la **gemcitabina.**

La miscela a "tre composti" è efficace negli esperimenti condotti sui topi





Una possibile nuova terapia per il mesotelioma?

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Dr. Annarita Miluzio

FONDAZIONE BUZZI UNICEM ONLUS
MESOTELIOMA MALIGNO

Prof. Bruno Burlando


2011
Nuclear Receptor Signaling Pathways


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