

# EFFECT ON THE CCRF-CEM CELLS EXPOSED TO RF/MW

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# **PROTOCOLLO SPERIMENTALE TEST MOBILE BADGE**

- **EXPOSURE IN TEM CELL of the cells CCRF-CEM by electromagnetic field of 900 MHz in presence and without MOBILE BADGE..**
- **900 MHz telephone connected to CMD 55**
- **PROLIFERATION TEST**
- **WB analysis CASPASI Bcl2**
- **FACS analysis**

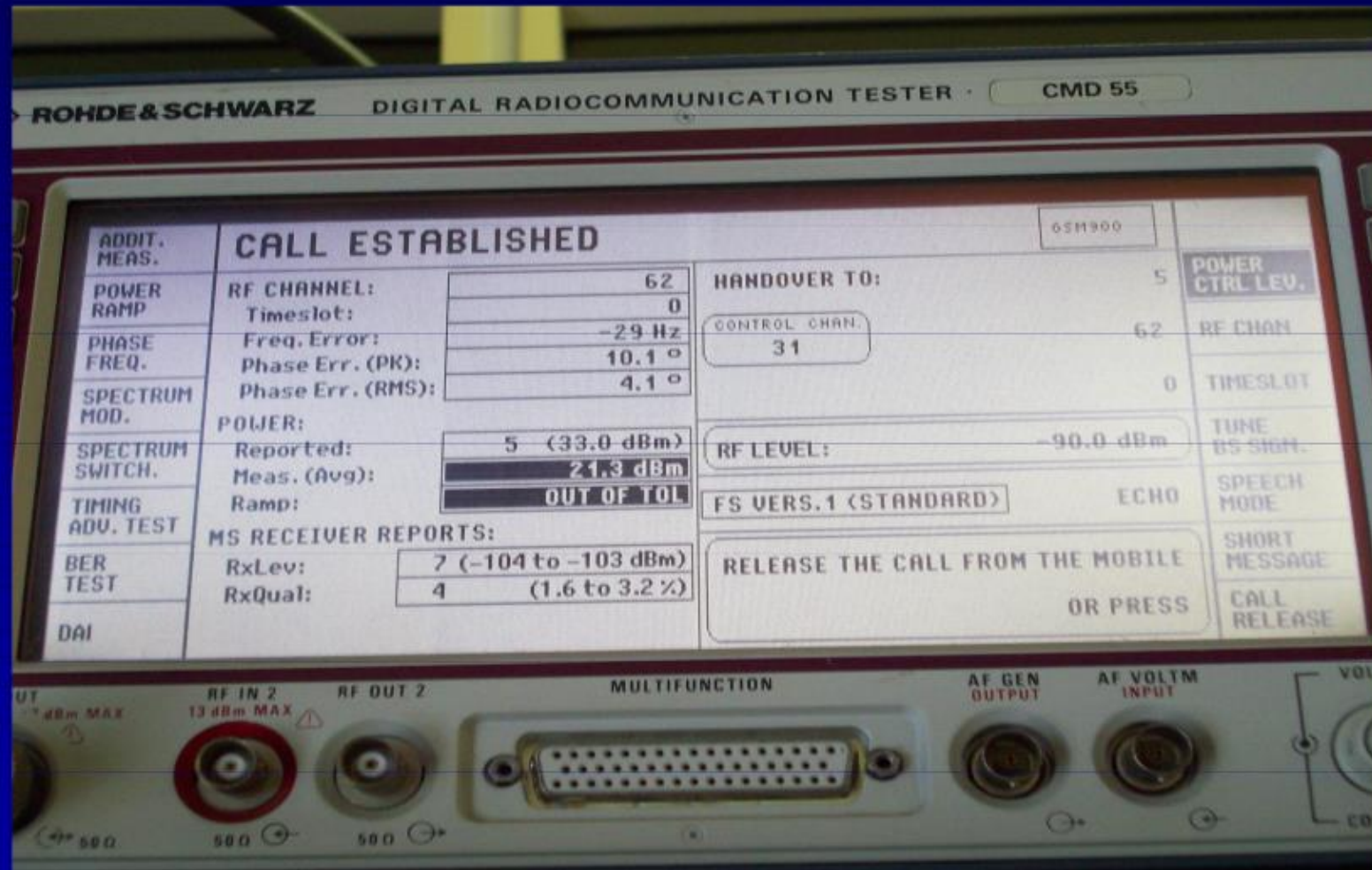
# GSM PER ESPOSIZIONI



# SEGNALE GSM 900 MHz IN CELLA TEM

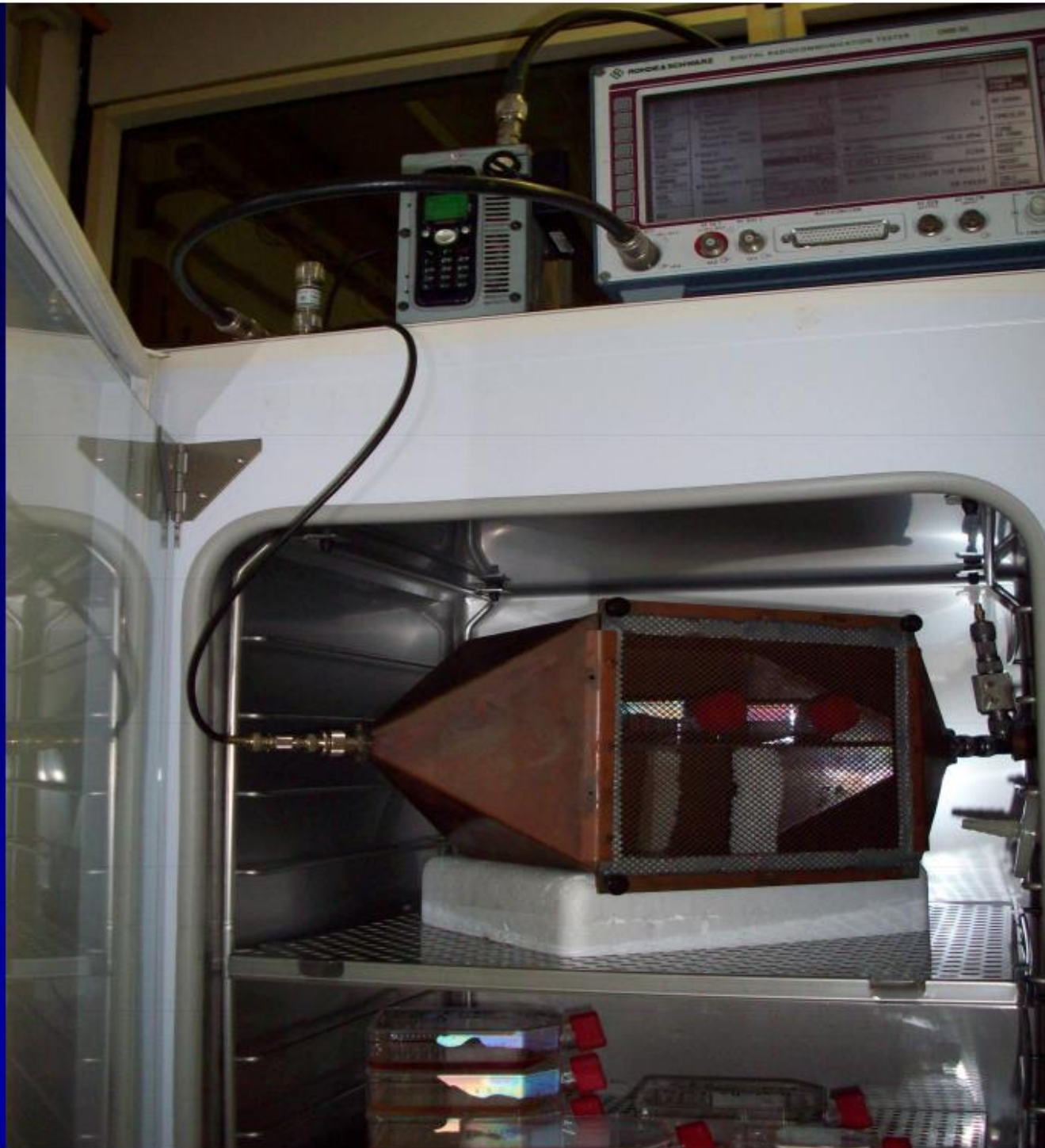


# CARATTERIZZAZIONE DEL SEGNALE GSM



# GSM -27.4 dBm

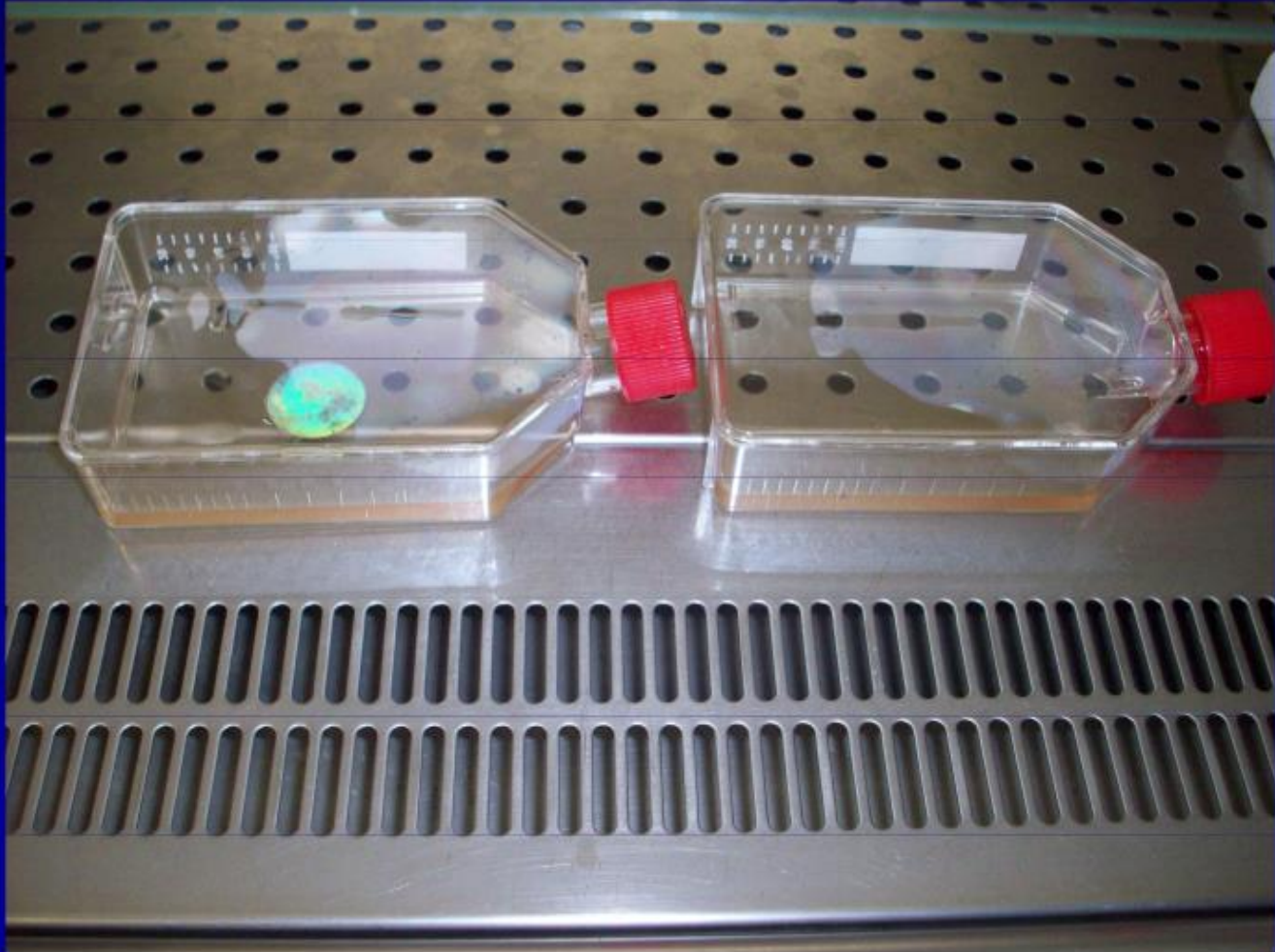






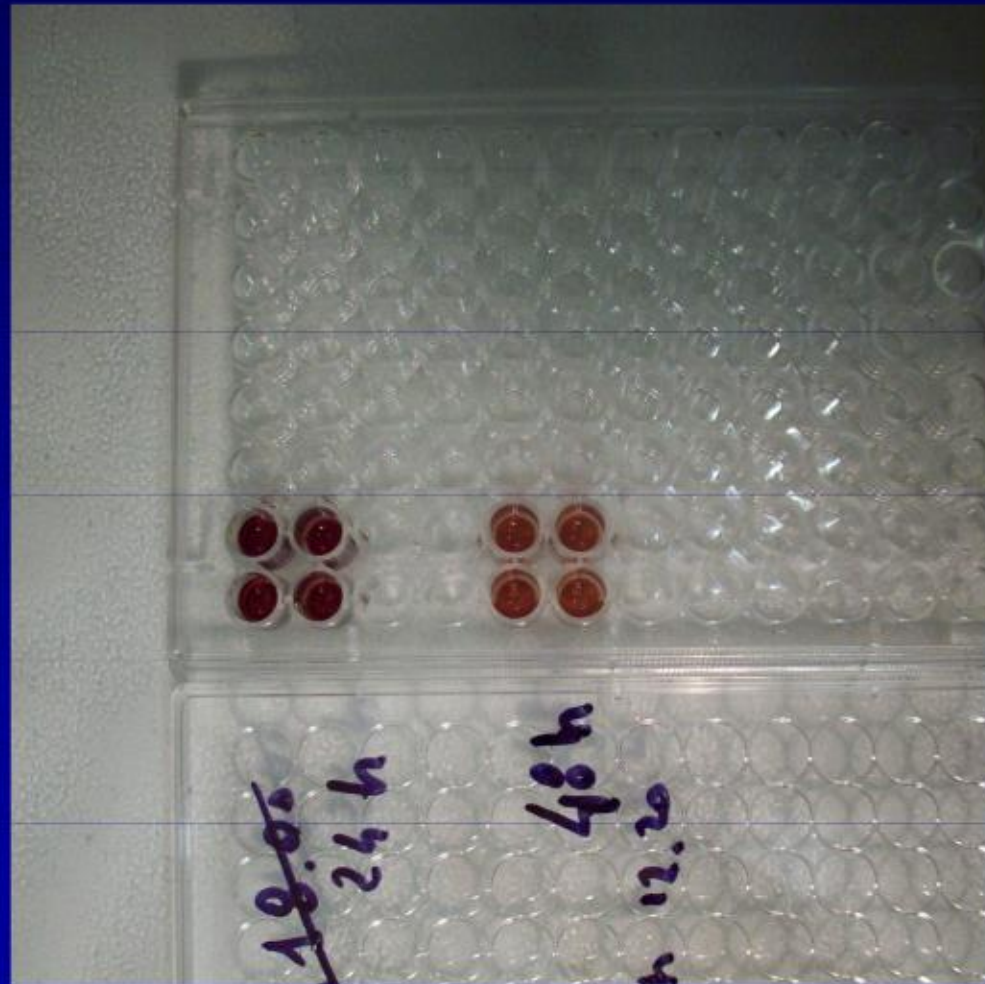








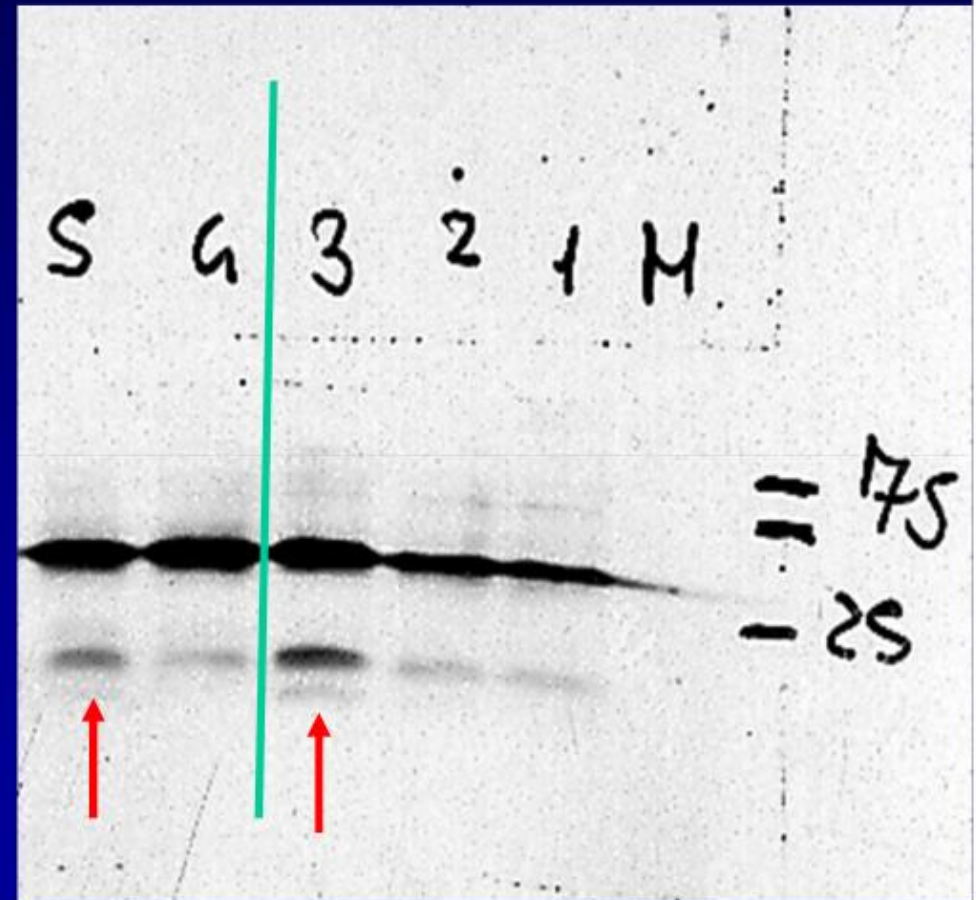
# PROLIFERAZIONE CELLULARE



# PRELIMINARY RESULTS WB

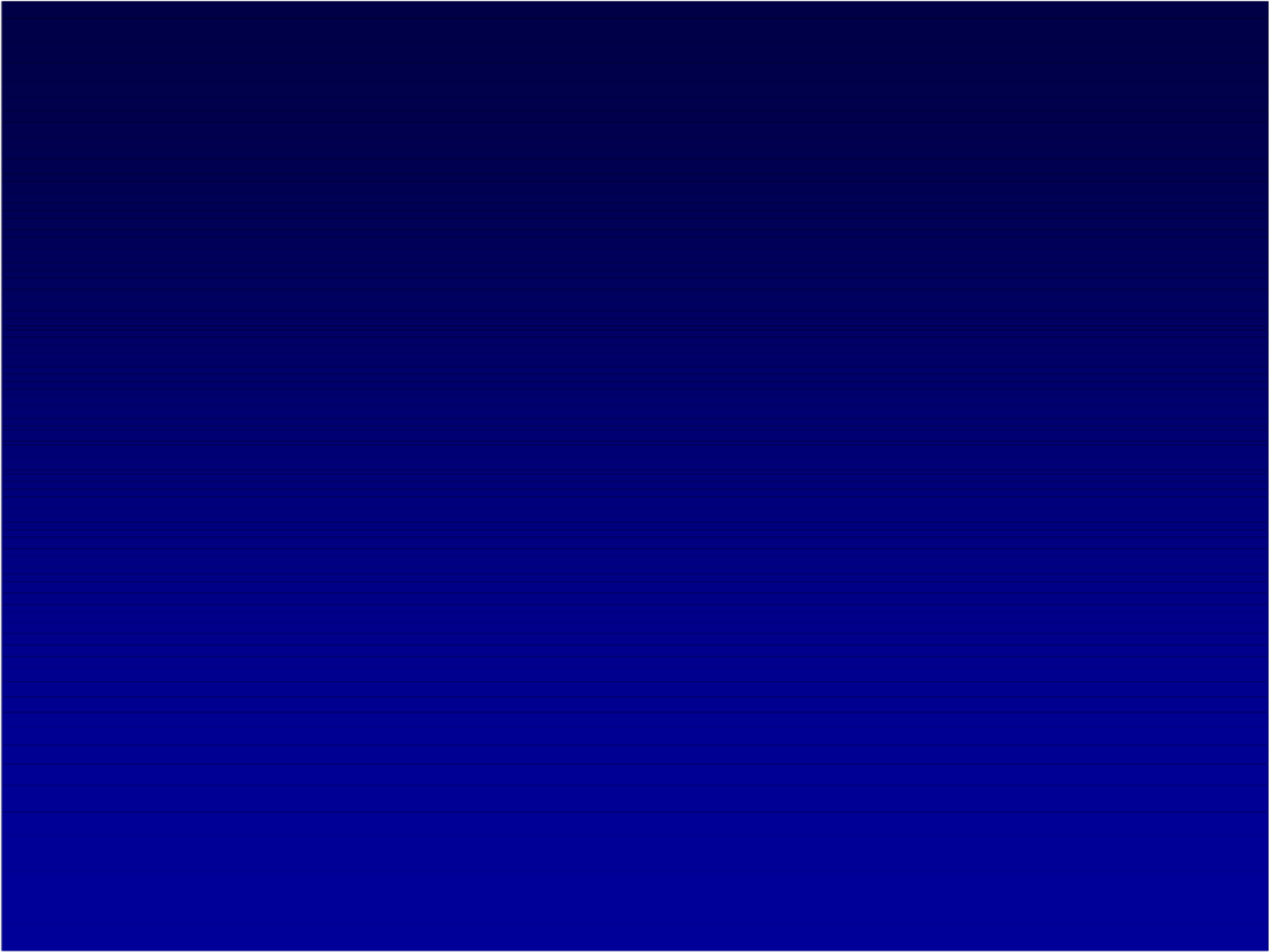
## Caspasi-3

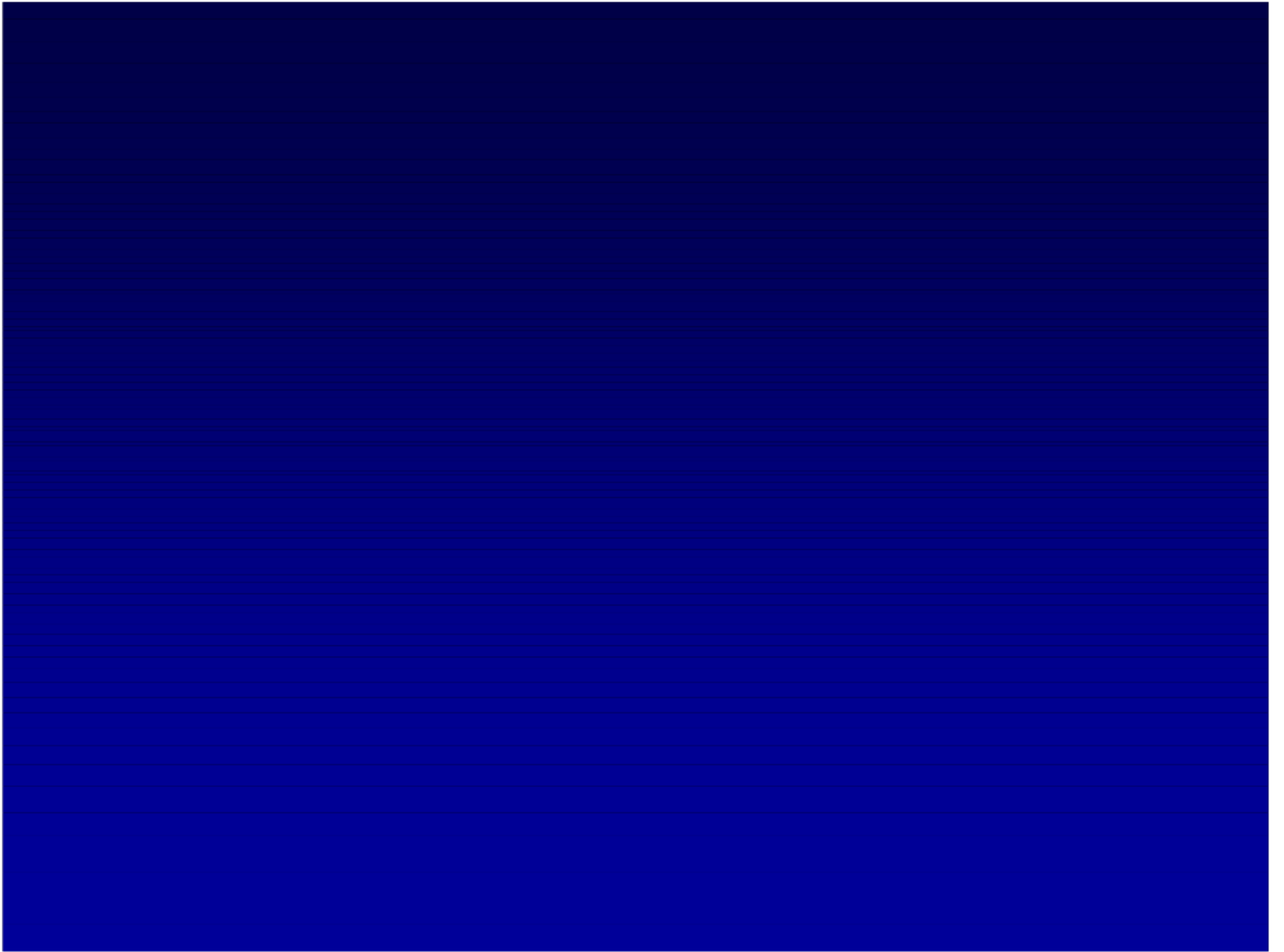
- 1- Ctrl 24h Out
- 2- E + B Exp 24h In
- 3- Exp 24h In
- 4- Ctrl 24h
- 5- Exp 24h



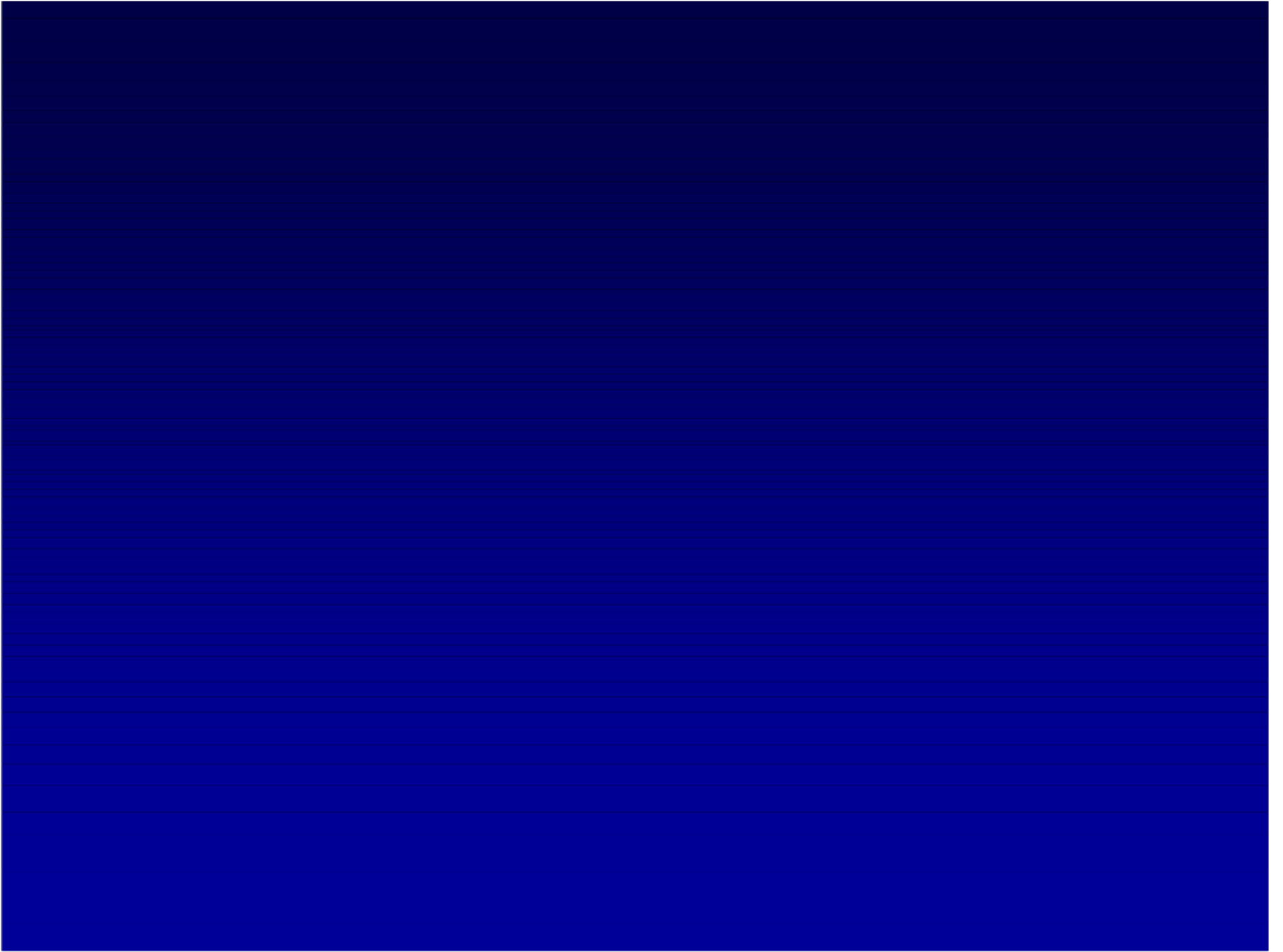
# Risultati I° Esperimento WB

- Effetto del Badge relativo alla espressione genica della Caspasi3 che induce apoptosi nelle cellule Esposte a Segnale GSM
- .
- Da testare l'antagonista BCL2
- Quantificare le proteine espresse.

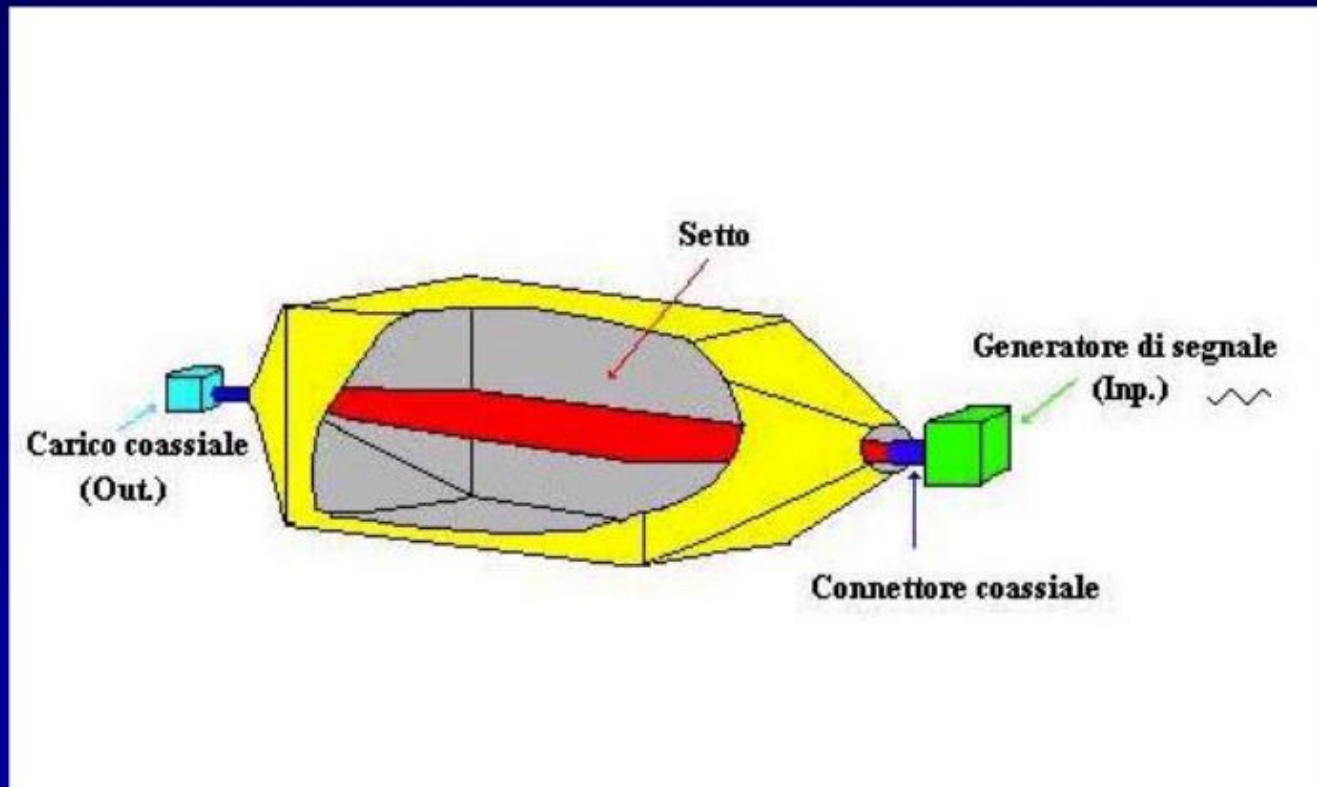








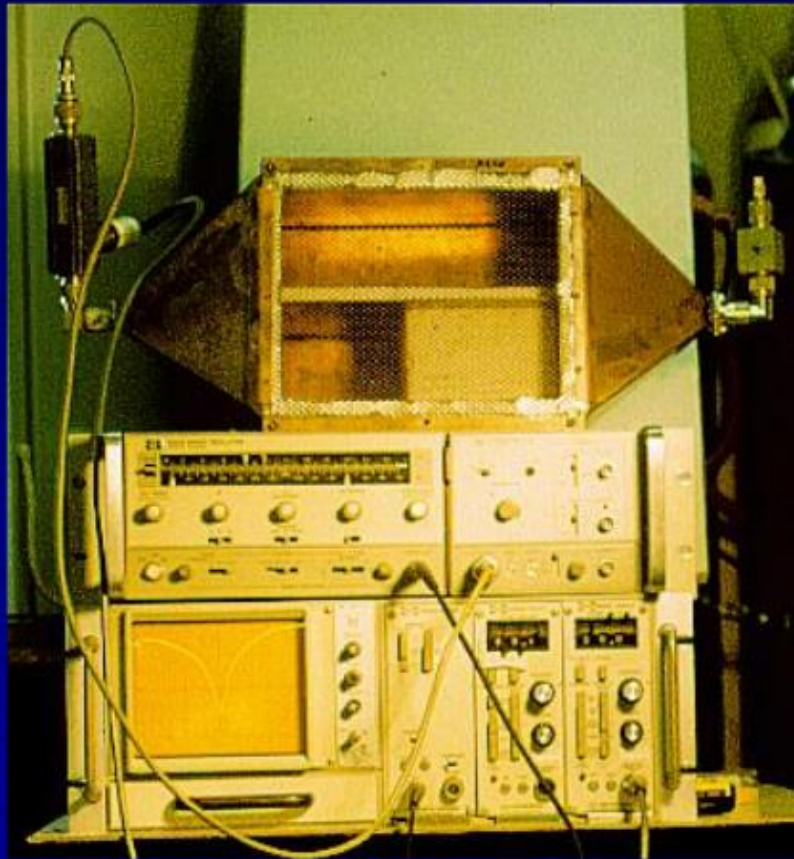
# EXPOSURE DEVICE TEM (transverse EM) CELL



# TEM CELL 900 MHz



# 800-1000 MHz EXPOSURE SYSTEM



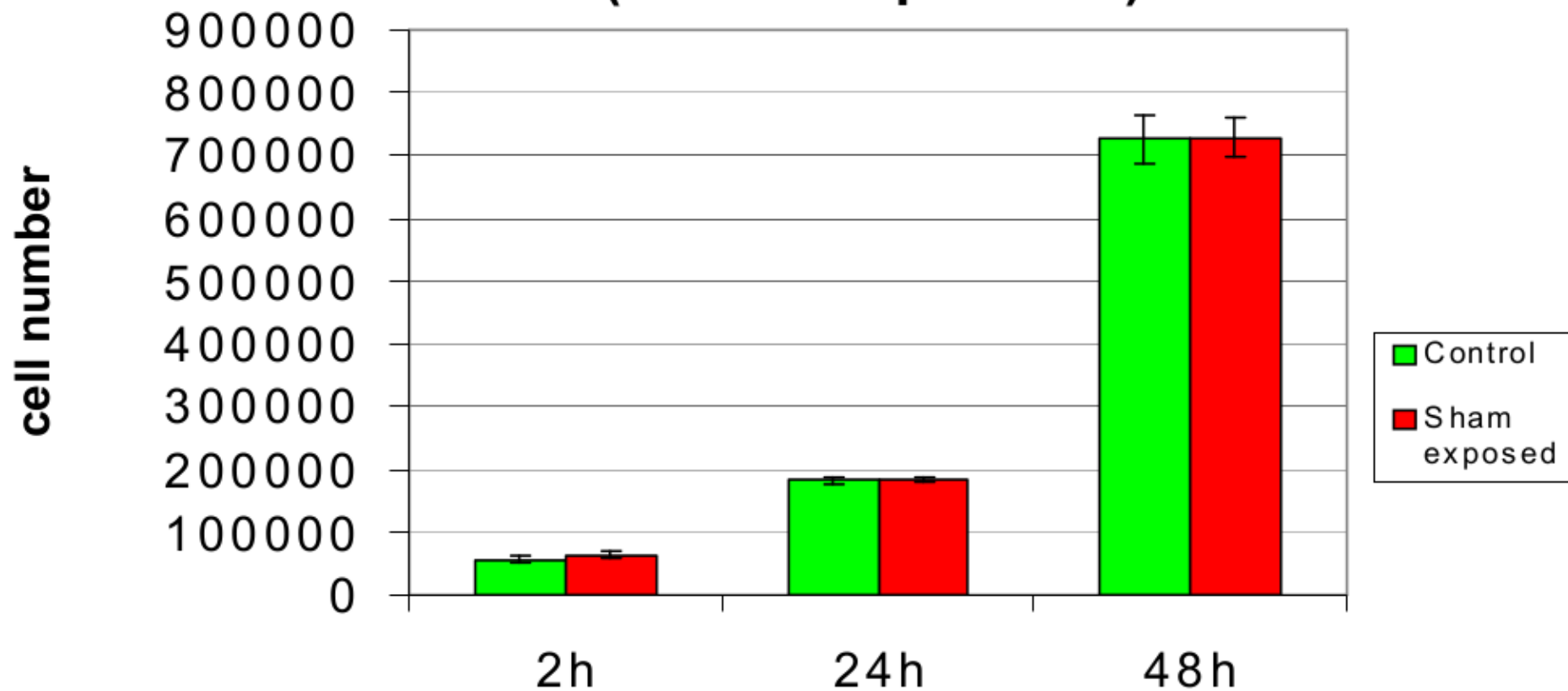
- Cella TEM
- Sweep Oscillator HP 8620 C
- Amplificatore HP 86222B RF  
10 - 2400 MHz
- Accoppiatore direzionale HP  
796D
- Testina bolometrica PM 10-  
0328
- Rivelatore PM 1038
- Universal counter HP 5316A
- Power meter HP 431 A

# TEMPERATURE CONTROL

<b>hours</b>	<b>T1 (CONTROL FLASK)</b>	<b>T2 (EXPOSED FLASK)</b>	<b>T3 (INCUBATOR)</b>
<b>2</b>	<b>37.01</b>	<b>37.01</b>	<b>37.00</b>
<b>24</b>	<b>37.00</b>	<b>37.00</b>	<b>37.00</b>
<b>48</b>	<b>37.00</b>	<b>37.00</b>	<b>37.00</b>

# SHAM EXPOSURE

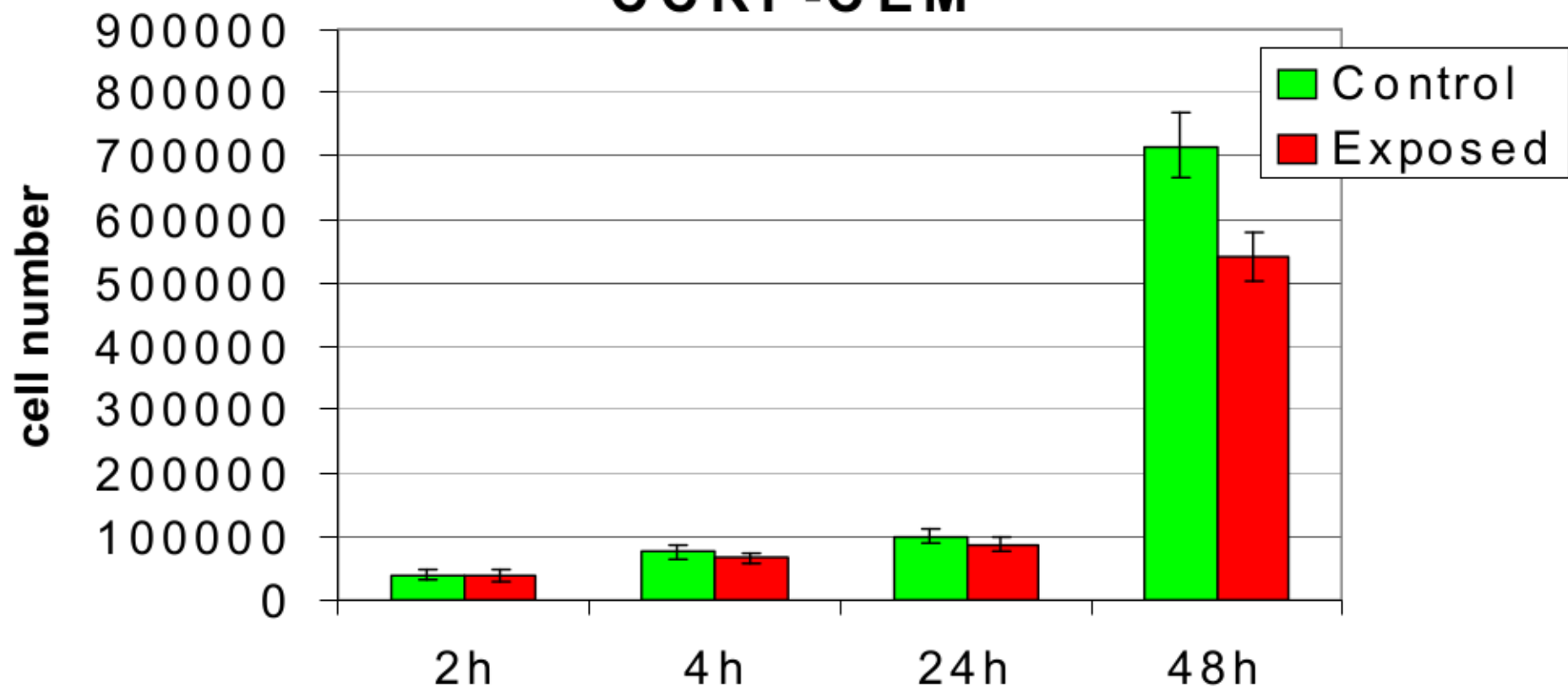
## Test di proliferazione in cellule CCRF-CEM (sham exposure)



# BIOLOGICAL EFFECTS

# PROLIFERATION TEST

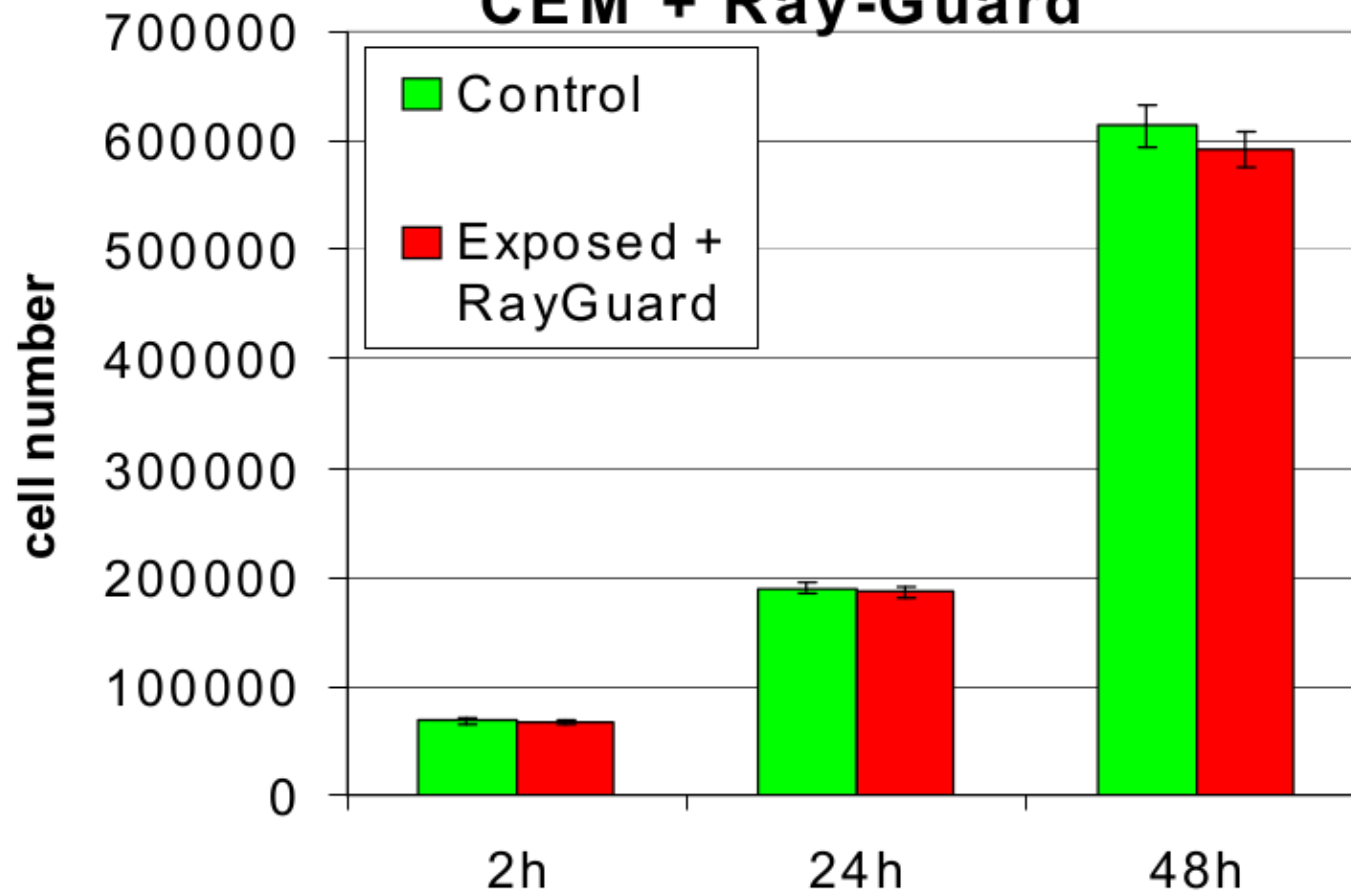
Test di proliferazione in cellule  
CCRF-CEM





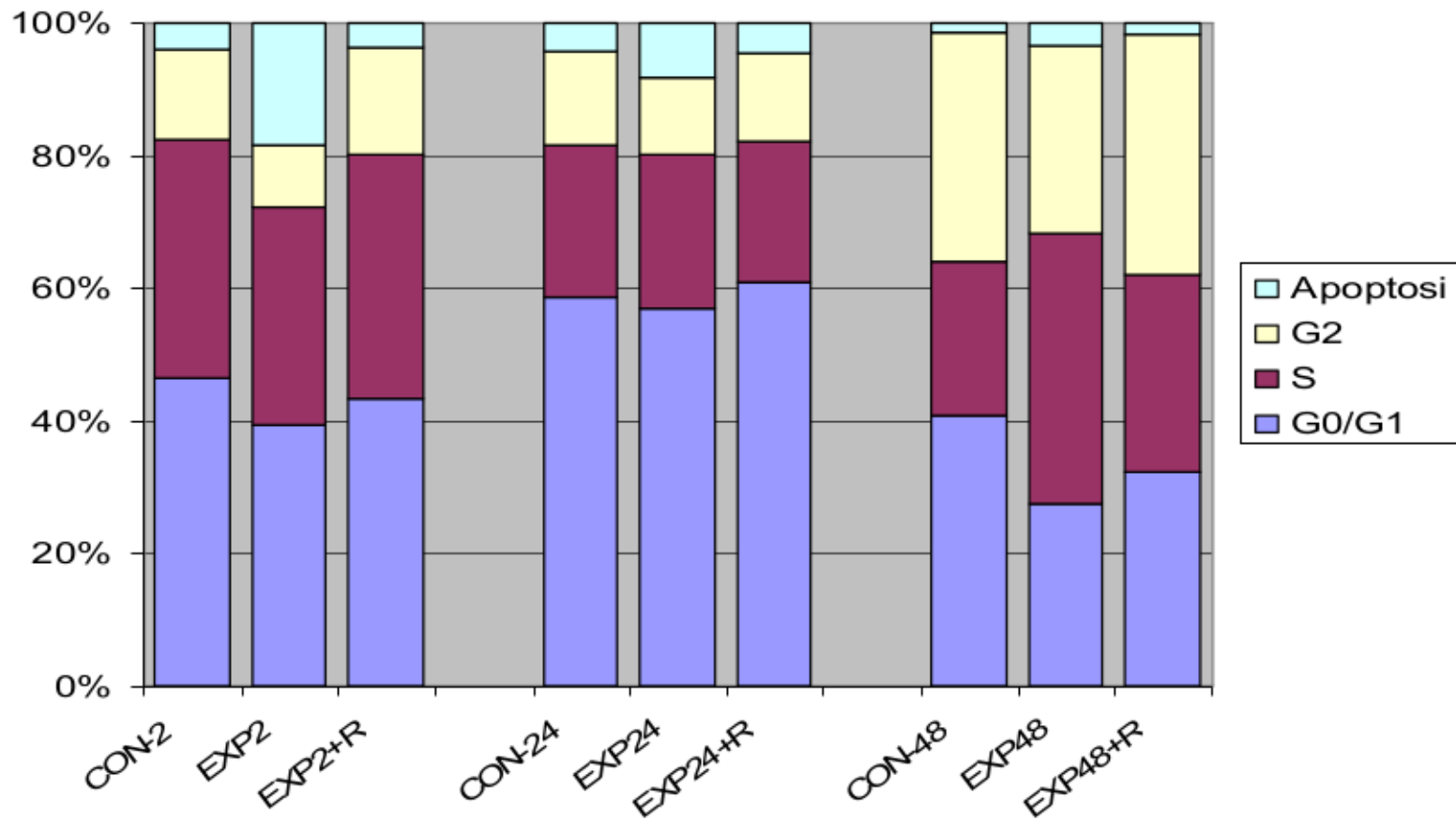
# EMF and RAY-GUARD

Test di proliferazione in cellule CCRF.  
CEM + Ray-Guard



# % OF CELL IN G0/G1, S, G2/M and APOPTOSIS IN CELL CYCLE PHASES

## CCRF-CEM FACS Analysis



# 2 HOURS 900 MHz EXPOSURE

	<b>G0/G1</b> (Quiescence)	<b>S</b> (DNA synthesis)	<b>G2/M</b> (mythosis)	<b>APOPTOSIS</b>
<b>CONTROL</b>	<b>45.75</b>	<b>35.55</b>	<b>13.42</b>	<b>3.89</b>
<b>NIR EXPOSED</b>	<b>38.5</b>	<b>32.38</b>	<b>9.13</b>	<b>18.07</b>
<b>NIR EXPOSED + RAY-GUARD</b>	<b>42.43</b>	<b>36.2</b>	<b>15.83</b>	<b>3.75</b>

# 24 HOURS 900 MHz EXPOSURE

	<b>G0/G1</b> (Quiescence)	<b>S</b> (DNA synthesis)	<b>G2/M</b> (mitosis)	<b>APOPTOSIS</b>
<b>CONTROL</b>	<b>56.31</b>	<b>21.95</b>	<b>13.79</b>	<b>4.3</b>
<b>NIR EXPOSED</b>	<b>54.82</b>	<b>22.15</b>	<b>11.09</b>	<b>7.89</b>
<b>NIR EXPOSED + RAY-GUARD</b>	<b>58.31</b>	<b>20.15</b>	<b>12.98</b>	<b>4.22</b>

# 48 HOURS 900 MHz EXPOSURE

	<b>G0/G1</b> (Quiescence)	<b>S</b> (DNA synthesis)	<b>G2/M</b> (mitosis)	<b>APOPTOSIS</b>
<b>CONTROL</b>	<b>40.05</b>	<b>22.60</b>	<b>33.97</b>	<b>1.37</b>
<b>NIR EXPOSED</b>	<b>26.68</b>	<b>39.83</b>	<b>27.62</b>	<b>3.38</b>
<b>NIR EXPOSED + RAY-GUARD</b>	<b>31.74</b>	<b>29.35</b>	<b>35.39</b>	<b>1.7</b>

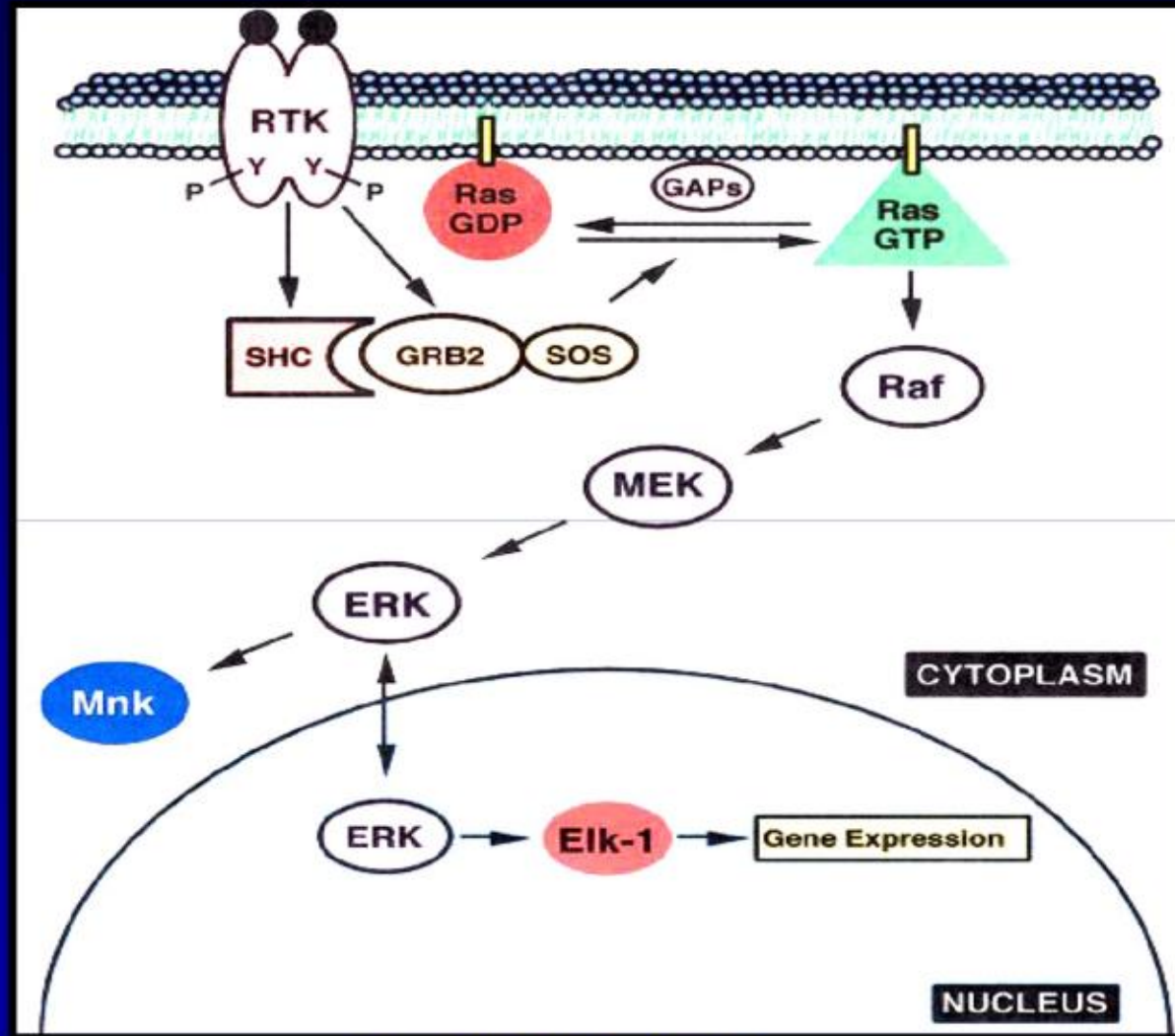
# CONCLUSIONS 1

- 1. Short exposure time (2 and 24 hours) at 900 MHz and 4.8 V/m electromagnetic fields induce apoptotic response in CEM cells. This effect has been strongly reduced by the presence of the RAY-GUARD device inside the TEM cell during exposure.**
- 2. Longer exposure time (48 hours) induces selection of tumorigenic clones, which show an higher level of DNA synthesis and a reduction of resting cells. The RAY-GUARD reduce the cycling cells and increase the resting quiescent cells. Moreover the apoptotic effect of NIR exposure is neutralized by the RAY-GUARD device**

## CONCLUSIONS 2

- 3. The biological effects induced by electromagnetic field are not related to the thermal effect.**
- 4. The electromagnetic fields can activate the cell cycle genes (i.e. RAS proto-oncogene) involved in the DNA synthesis control.**
- 5. Should their expression be controlled by RAY-GUARD?**

# RAS SIGNALING



from Vojtek AB and Der CJ,  
JBC 273 n.32:19925-19928



# WB ANALYSIS

## H-RAS EXPRESSION

- **1,3,5,7 SAMPLES=**  
**CONTROL CELLS**  
**2,4,24,48 hs**



- **2,4,6,8 SAMPLES=**  
**EXPOSED CELLS**  
**2,4,24,48 hs**

<b>Power Density</b>	<b>Reported Biological Effects</b>	<b>References</b>
0.168 - 1.053 $\mu\text{W}/\text{cm}^2$	Irreversible infertility in mice after 5 generations of exposure to RFR from "antenna park"	Magras & Xenos, 1997
0.16 $\mu\text{W}/\text{cm}^2$	Motor function, memory and attention of school children affected (Latvia)	Kolodynski, 1996
0.2 - 8 $\mu\text{W}/\text{cm}^2$	Two-fold increase in childhood leukemia / RFR exposure to AM/FM towers	Hocking, 1996
1.0 $\mu\text{W}/\text{cm}^2$	Whole body microwave irradiation of male mice caused a significant effect on the immune system	Fesenko, 1999
1.0 $\mu\text{W}/\text{cm}^2$	Irradiation (5 hours) with low-power microwaves stimulates the immune potential of macrophages and T cells	Novoselova, 1999
1.3 - 5.7 $\mu\text{W}/\text{cm}^2$	Two-fold increase in leukemia in adults from AM RF exposure	Dolk, 1997
~2-4 $\mu\text{W}/\text{cm}^2$	Direct effect of RFR on ion channels in cells/opening of acetylcholine channels	D'Inzeo, 1988
4-10 $\mu\text{W}/\text{cm}^2$	Visual reaction time in children is slowed//lower memory function in tests	Chiang, 1989
5 - 10 $\mu\text{W}/\text{cm}^2$	Impaired nervous system activity	Dumansky, 1974
10 $\mu\text{W}/\text{cm}^2$ (0.0027 W/Kg SAR)	Changes in active avoidance conditioned reflex (behavioral change) after 0.5 hour exposure	Navakatikian, 1994
10-20 $\mu\text{W}/\text{cm}^2$	Increase in micronuclei (abberant DNA form) found in workers chronically exposed to microwaves at 1250-1350 MHz.	Garaj-Vrhovac, 1995
10 - 25 $\mu\text{W}/\text{cm}^2$	Changes in the hippocampus of the brain	Belokrinitsky, 1982
30 $\mu\text{W}/\text{cm}^2$ (0.015 W/Kg SAR)	Immune system effects - elevation of PFC count (antibody-producing cells)	Veyret, 1991
50 $\mu\text{W}/\text{cm}^2$	An 18% reduction in REM sleep (important to memory and learning functions)	Mann, 1996
100 $\mu\text{W}/\text{cm}^2$	Changes in immune system function	Elekes, 1996
100 $\mu\text{W}/\text{cm}^2$ (0.027 W/Kg SAR)	A 24% drop in testosterone after 6 hours exposure	Navakatikian, 1994

# THE RESEARCH GROUP

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# REMARKS

- **It is hardly possible to transfer these results automatically to humans.**
- **We do not know if the Ray-Guard can protect efficiently the organisms.**
  - **We suppose it! As we obtained preliminary results on human lymphocytes**

# New experimental aims on normal lymphocytes

1. Exposure of healthy donor lymphocytes to RF/MW 900 MHz in presence of Ray-Guard device.
2. Analysis of the electromagnetic field effect on cell cycle in presence of Ray-Guard device.

## New proposal research

- Ray-Guard effect on modulation of cell cycle genes expression in CCRF-CEM cells exposed to NIR, such as oncogenes (i.e.Ras) and onco-suppressor genes.