



## R.G.C.C.-RESEARCH GENETIC CANCER CENTRE ltd

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we send you the results from the analysis made about a patient suffering from squamous cell carcinoma stage ? The sample that was sent to us for analysis was a sample of 40ml of whole blood that contained EDTA-Ca as anti-coagulant , packed with water ice .

In our laboratory we made the following :

- We isolated the malignant cells using Oncoquick with a membrane that isolates malignant cells from normal cells . Then we centrifuged at 350g for 10 min and we collected the supernatant with the malignant cells . Then we proceed to isolation of malignant cells from mononuclear cells by negative selection .
- Then we developed thirty eight cell cultures in a fetal calf serum media . In each culture of the well plate we added a biological modifier substance (H<sub>2</sub>O<sub>2</sub>, ascorbic acid , carnivora , misteltoe, quercetin , indol-3-carbinol , c-statin , ukrain , polyMVA, co enzyme Q10, essiac tea, modified citrus pectin, IP6 , pancreatic enzymes, salvestrol, Uncaria Tomentosa, carctol, noni juice, annonaceous acetogenins, caesium chloride, reolysin, amygdalin-B17-, artesunate, maitake, lycopene, curcumin, green tee extract, melatonin, ellagic acid, L-methionine, N-acetyl-cystein, Niacin (Vit.B3), L-carnithine, Vitamin E (tocopherol), superoxide dismutase (SOD) , selenium, aloe vera, IFN $\alpha$ 2) that is used in clinical application. Then we developed those cultures and we harvested a sample every 24 hours and made the following assays.
- In the culture that it contains all substance we measure the apoptotic ability using the oncogen apoptosis kit
- In the culture that it contains the carnivora we measure the inhibition of tyrosin kinase catalytic ability from growth factors receptor (EGF-r, IGF-r,) and the production of cytokines.
- In the culture that it contains the Ukrain we measure the inhibition of tyrosin kinase catalytic ability from growth factors receptor (EGF-r, IGF-r,) and the production of cytokines PMBC
- In the culture that contains quercetin we measure the inhibition of EGF and IGF .
- In the culture that contains indol-3-carbinol we measure the inhibition of VEGF and FGF and PDGF
- In the culture that it contains the misteltoe we measure the inhibition of tyrosin kinase catalytic ability from growth factors receptor (EGF-r, IGF-r,) and the production of cytokines and the increase of PMBC
- In the culture that it contains the H<sub>2</sub>O<sub>2</sub> we measure viability of the culture in 4 days of treatment.
- In the culture that it contains the ascorbic acid we measure the catalytic activity of GSH and GSSG (redox reaction) and the induction of cytochrome C (apoptosis).
- In the culture that it contains the PolyMVA we measure the catalytic activity of GSH and GSSG (redox reaction) and the induction of cytochrome C (apoptosis)
- In the culture that it contains the artesunate we measure the catalytic activity of GSH and GSSG (redox reaction for free radical since artesunate bind free radicals with iron molecule ) , the inhibition of VEGF , FGF and PDGF



(since it act to the angiogenesis cascade reactions) and the induction of cytochrome C (apoptosis).

## RESULTS:

1. We notice that in culture that contains the ascorbic acid we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by 50%.
2. We notice that in culture that contains the PolyMVA we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by 20%.
3. We notice that in culture that contains carnivora we have inhibition of EGF-r by less than 5% and for IGF-r by 5% and we notice no increase of cytokine production 5% .
4. We notice that in the culture that contains quercetin we have inhibition of EGF by 60% and IGF by 45%
5. We notice that in the culture that contains indol-3-carbinol we have inhibition of VEGF by less than 5% , of FGF by 5% , and PDGF by 5%
6. We notice that in culture that contains misteltoe we have inhibition of EGF-r by 55% and for IGF-r by 35% and we notice increase of cytokine production by 50%, and there is increase of PMBC by 30%.
7. We notice that in culture that contains the c-statin we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by 55%.
8. We notice that in culture that contains Ukrain we have inhibition of EGF-r by less than 5% and for IGF-r by 5% and we notice no increase of cytokine production , and there is no increase of PMBC .
9. We notice that in culture that contains the H2O2 we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable .
10. We notice that in culture that contains the co enzyme Q10 we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable .
11. We notice that in culture that contains the essiac tea we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable .
12. We notice that in culture that contains the modified citrus pectin we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable .
13. We notice that in culture that contains the IP6 we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable .
14. We notice that in culture that contains the pancreatic enzymes we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable .
15. We notice that in culture that contains the salvestrol we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
16. We notice that in culture that contains the uncaria tomentosa we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable.
17. We notice that in culture that contains the caesium chloride we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.



18. We notice that in culture that contains the carotol we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable.
19. We notice that in culture that contains the noni juice we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable.
20. We notice that in culture that contains the annonaceous acetogenins we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable.
21. We notice that in culture that contains the reolysin we have increase of the cascade of caspase (especially 3 and 9) and cytochrom-c by less than 5% and the viability of the culture remain stable.
22. We notice that in culture that contains the amygdalin-B17- we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
23. We notice that in culture that contains maitake we have inhibition of EGF-r by less than 5% and for IGF-r by 5% and we notice no increase of cytokine production , and there is no increase of PMBC.
24. We notice that in culture that contains the curcumin (turmeric) we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
25. We notice that in culture that contains the lycopene we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
26. We notice that in culture that contains the green tea extract we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable
27. We notice that in culture that contains artesunate , there is inhibition of redox reaction and no increase of intracellular free radicals , there is increase of cytochrome c (apoptosis) by 50% and the inhibition rate of VEGF is 55% , of FGF is 35% and of PDGF is 35%.
28. We notice that in culture that contains the melatonin we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
29. We notice that in culture that contains the ellagic acid we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
30. We notice that in culture that contains the L-methionine we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
31. We notice that in culture that contains the N-acetyl-cystein we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
32. We notice that in culture that contains the niacin we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
33. We notice that in culture that contains the L-carnithine we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
34. We notice that in culture that contains the vitamin E we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.

35. We notice that in culture that contains the superoxide dismutase we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
36. We notice that in culture that contains the aloe vera extract we have no increase of the cascade of caspase (especially 3 and 9) and cytochrom-c and the viability of the culture remain stable.
37. We notice that in culture that contains selenium we have inhibition of EGF-r by less than 5% and for IGF-r by 5% and we notice no increase of cytokine production , and there is no increase of PBMC and NK .
38. We notice that in culture that contains IFNa2 we have inhibition of EGF-r by less than 5% and for IGF-r by 5% and we notice no increase of cytokine production , and there is no increase of PMBC .

**CONCLUSION:** It seems that this specific population of malignant cell have greater sensitivity in quercetin, in ascorbic acid, in Poly-MVA, in artesunate, in misteltoe and in c-statin and less in hydrogen peroxide (H2O2), co enzyme Q10, essiac tea, modified citrus pectin, IP6 , N-acetyl-cystein, pancreatic enzymes, caesium chloride, in carnivora, ellagic acid, in L-carnithine, in L-methionine, vitamin E (tocopherol), maitake, in IFNa2, in amygdalin-(B17), in curcumin (turmeric), in indol – 3-carbinol, uncaria tomentosa (samento), in melatonin, in selenium, caretol, in ukrain, noni juice, niacin, aloe vera extract, in superoxide dismutase, in salvestrol, annonaceous acetogenins (paw paw), reolysin (reovirus), lykopen and in green tea extract .

Regardsly,



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