A New Alternative Medicine can cause Selective Cell Death in Cancer Cells

Principle investigator:

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Research team:

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Collaborators:

Abdel Moneim Osman – Medical Pharmacy, Awatif Mohammad – Medical biology, Hanan Al Shathli- Medicine , Manal Khorshid- Medicine , Nadiah Spki – Medicine, Zuhoor Al Gheithi – Medicine, Sami Alnuaim – Medicine, Alaa Keder - Pharmacology, Mahmoud Shahin- Oncology, Abdulwahab Badr - Dermatology, Samiha Al Shathli- Dermatology, Fadwa Khorshid-Medicine,Haifaa Janna-Dermatology and others.

Introduction:

قال تعالى: (أَفَلَا يَنظُرُونَ إِلَى ٱلْإِبِلِ كَيْفَ خُلِقَتْ)"سورة الغاشية"

Do they not look at the camels, how they are created?

روى البخاري عن أنس أن رهطاً من المدينة قدموا على النبي صلى الله عليه وسلم فقالوا: إنا اجتوينا المدينة فعظمت بطوننا وارتهشت اعضادنا فأمرهم النبي صلى الله عليه وسلم أن يلحقوا براعي الإبل فيشربوا من ألبانها وأبوالها حتى صلحت بطونهم وألوانهم...

The saheeh hadeeth says that some people came to Madeenah then fell sick with huge abdomens. The Prophet (peace be upon him) told them to drink the milk and urine of camels, they recovered and grew fat. Narrated by Al-Bukhaari (2855) and Muslim (1671). With regard to the health benefits of drinking the milk and urine of camels, they are many, well known to the earlier generations of medical science and they have been proven by modern scientific researches.

Swollen Abdomen may indicate oedema and liver disease (jaundice) or cancer, and thin bodies demonstrated the extreme weakness, which often accompanies with

hepatitis or cancer….This was a beginning of long and arduous way, but it is full of light, and prosperity from Allah.
We thank god and appreciated to every one who contributed money or effort or advice to this work.

Ethical Approval:

The research approved from the ethics committee of scientific research, King Abdul Aziz University at:

- **2/3/2005 to apply to the cells and animal models.**
- **1/16/2008 to apply to human- healthy.**
- **28/10/2009 to apply to human-patients.**

Funds & Prizes:

- **The research studied the effect of substrate from the prophet medicine (camels urine), which coded as PM701.**
- **The actual work started of about 2004, then the research funded by KAU for 5 times with (014/425 two phases); (001/427 two phases); (02n/427 one phase).**
- **The research obtained 1st Position in Al-Ibtikar exhibition of Saudi Arabia Inventors at Ibtikar 2008.**
- **The research funded by ALZAMEL as a Scientific Chair no "429/3/KBM" at 20/6/2008.**
- **ITEX GOLD Medal at the 20th International Invention, Innovation & Technology Exhibition ITEX 2009, Kuala Lumpur, Malaysia , held from 15th - 17th May 2009.**
- **Awarded the ASIA CUP 2009 FINALIST at the 20th International Invention, Innovation & Technology Exhibition ITEX 2009, Kuala Lumpur, Malaysia held from 15th - 17th May 2009.**
- **Honor Award from Prince Mishaal bin Abdul Aziz in 30/ 1/ 2010.**

Patents:

- **Patent recorded in GCC no.9962 at 16/1/2008.**
- **Patent recorded in US Patent No. 12/178,152 at 23/7/2008.**
- **Patent recorded in Europe Patent Office (EPO), No.09162954.3 at 17/6/2009.**
- **Patent recorded in Chinese Patent office (SIPO), No. 200910168793.7 at 3/9/2009.**

Summery:

Camel urine or PM 701 was clean, sterile and free from toxicity when it was studied by our team work. We used PM 701 as anticancer agent. The work was conducted as in vitro, in vivo, preclinical and clinical studies.

The study used the technique of tissue culture of human lung cancer cells and mice leukemia cells, which were compared to with normal human skin fibroblasts in studying the effect of PM 701. This proved that our agent could induce apoptosis of the cancer cells without affecting the normal cells. The in vivo studies tested the effect of PM 701 in treating MFI mice, which were inoculated with L1210 cells. The result of in vivo is satisfactory as in vitro, where the life span and the survival of animals improved. The histopathology studies revealed disappearance of malignant tumors in the organs of treated animals or at least a decrease the in the degree of metastasis. So PM 701 showed promising results as anticancer agent in cancer induced animal model. For enhancing the utility and convenience of application of PM701, liquid PM701 was lyophilized to reach a solid form. Then the solid form was fractionated to obtain the bioactive fraction(s), which were more readily acceptable to humans. During these steps, we monitored the activity of the different fractions using the different tests subjected previously on the crude PM 701, such as in vitro and in vivo tests. The fraction, which was coded PMF (150mg/g of the lyophilized PM701) was able to significantly inhibit the proliferation of cancer cells without affecting the normal cells at cell culture levels. So PMF is the ideal selective cytotoxic agent whereas the other fractions are less effective or non-selective. On further fractionation, PMF was subfractionated to seven parts in order to isolate the most effective cytotoxic constituents of this bio-active fraction. Our studies identified that the subfraction PMFK was the most cytotoxic constituents. PMF and PMFK had been examined and showed successful results in preventing the growth of cultivated cancer cells for different carcinogenic cell lines as in the liver, colon, brain and breast in addition to the lung and leukemia.

The pharmacological and toxicological studies were done on formulated PM701 and PMF on animal models. These experiments proved their safety as indicated by the high value of MTD on animal models and also by its safety effect on vital organs of animal models through histopathological studies carried out using light and electron microcopies.

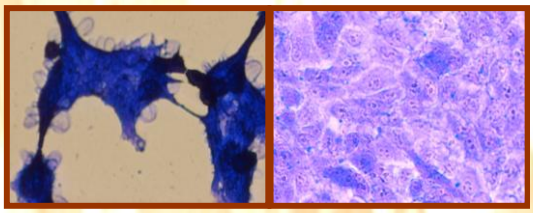
The clinical trail on human indicated that the application on healthy volunteers using the capsules and syrup containing PMF did not appear to

have any harmful side effects. Whereas the size of the tumor was reduced 50% in a third degree patient volunteer with lung cancer, after use of medication for only one month, reduced 80% after two months and there is no active cells after 9 months. In other patient (pancreatic cancer without any other treatment) the biomarker (CA99) reduced from 9600 to 2500 after only three weeks. In another metastatic patient, the biomarker (CEA) reduced from 294 to 194 in two weeks and the tumor in liver reduced 2mm. The treatments are still ongoing for all these patients.

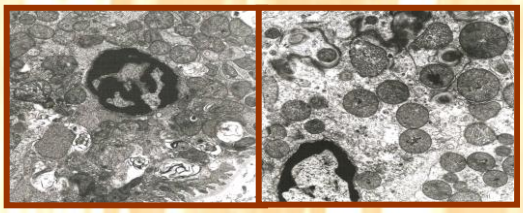
Publications:

1. Khorshid FA, Moshref SS, Heffny N. An Ideal Selective Anticancer Agent In Vitro, I- Tissue Culture Study of Human Lung Cancer Cells A549. JKAU- Medical Sciences .Vol 12, PP 3-18, 2005.
2. Moshref SS, Khorshid FA, Jamal Y. The Effect of PM 701 on Mice Leukemic Cells:I - Tissue Culture Study of L1210 (In Vitro) II - In Vivo Study on Mice, JKAU- Medical Sciences .Vol 13 (1), pp. 3-19, 2006.
3. Khorshid FA. Moshref SS. In Vitro Anticancer Agent, I - Tissue Culture Study of Human Lung Cancer Cells A549 II - Tissue Culture Study of Mice Leukemia Cells L1210. International Journal of Cancer Research 2 (4):330-344, 2006.
4. Khorshid FA. Preclinical evaluation of PM 701 in Experimental animals. International Journal of Pharmacology, 4(6): 443-451, 2008. ISSN 1811-7775.
5. Moshref SS PM701 A Highly Selective Anti Cancerous Agent Against L1210 Leukemic Cells: II – In Vivo Clinical And Histopathological Study. JKAU- Medical Sciences .Vol 14 (1), pp.85-99, 2007.
6. Khorshid FA. Potential Anticancer Natural Product against Human Lung Cancer Cells. Trends in Medical Research 4 (1): 9-15, 2009.
7. Raouf GA, Khorshid FA; Kumosani T. FT-IR Spectroscopy as a Tool for Identification of Apoptosis-Induced Structural Changes in A549 Cells Dry Samples Treated with PM 701. Int. J. Nano and Biomaterials, Vol. 2, No. 1/2/3/4/5, 2009
8. Khorshid FA. Osman AA. Abdulsattar E. Cytotoxicity of Bioactive fractions from PM 701. EJEAFChe, 8 (11), 2009. [1091-1098].
9. El-Shahawy A, El-Sawi N., Backer W.S. , Khorshid F.A., and Geweely N.S. Spectral Analysis, Molecular Orbital Calculations And Antimicrobial Activity Of PMF-G Fraction Extracted From PM-701. Int. J. of Pharma and Bioscience, vol. 1(2): p 1-19, 2010
10. Khorshid F.A., Shazly H., Al-Jefery A., and Osman A.A. Dose Escalation Phase I Study in Healthy Volunteers to Evaluate the Safety of a natural product PM 701. Int. J. of pharmacology and toxicology, 5(3): 91-9,2010

Culture Cell *(in vitro)*

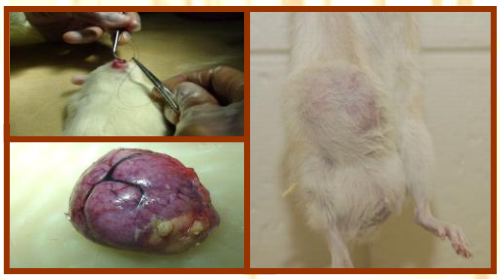


Treated cancer cells A549 after 24 h (a) with PM 701, note the damage of cells as compared with non-treated cells (b).



EM showed the apoptosis, as shown by chromatin condensation and membrane blabbing in treated cells with PM701

Animal experiments *(in vivo)*



Orthotropic surgery of Rat with human melanoma tissues showed subcutaneous swelling mass in the right inguinal region of the rat after one year of transplantation. Note the removal mass in left lower picture.

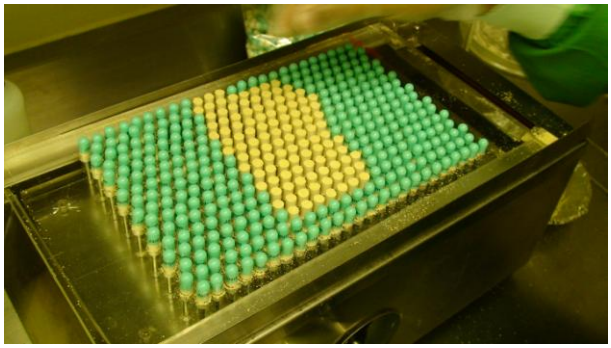
Our products under investigation which must used under medical observation

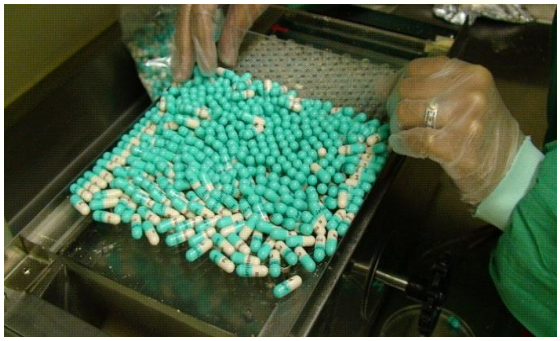
Manufactured after the adoption of Al-Zamil Chair for cancer research – No MBK/3/429 in 20 / 6 / 2008 for a period of three years.

Ointment: is used for burns caused by radiotherapy and the majority of skin diseases and eczema.

Lotion: is used for burns caused by radiotherapy and the majority of skin diseases and eczema.

Capsules: are under test for carcinogenic diseases and each capsule contain 300 mg of active substance PMF.





Syrup: is under test for carcinogenic diseases and each tea spoonful (5ml) contain 1000 mg of active substance PMF.