國立台灣大學電機資訊學院　電機工程學研究所

博士論文

Department of Electrical Engineering

College of Electrical Engineering and Computer Science

National Taiwan University

Doctoral Dissertation

人類健康及其風險之免疫發炎模型

AN IMMUNE-INFLAMMATORY MODEL OF HUMAN LONGEVITY AND RISKS

王正邦

Jeng-Pang Wang

指導教授：張璞曾 教授

共同指導：林耀仁 教授

Advisor: Fok-Ching Chong, Professor

Yaw-Jen Lin, Professor

中華民國 98 年 7 月

July, 2009

ACKNOWLEDGMENTS

First of all, I would like to thank my advisor Professor Fok Chin Chong. His wisdom , his mercy , his kindness has guided me into a new field of study, biosignal, and a new level of life philosophy. He is always willing to help not only his acquaintance or relatives, but also a lot of people in suffering, in illness . Help them away from bitterness and toward happiness. He is always my mentor, and will be my role model forever.

I want to appreciate Professor Chun Den Tseng for his encourage and permission to let me go to the graduate school, for his advise on cardiology, and for his help about my being visiting-scholar to the Queensland University with Professor Daniel Hung.

Specially, I express my gratitude to Professor Shiu-Nan Chen for his advise and help in immunotherapy and cardiovascular study. He overwhelms me with his enthusiasm toward innovation and contribution in helping cancer and cardiovascular patients.

And I would like to set my best regards to professor Yaw Jen Lin and his family, not just for his being my advisor in medical informatics but also for their tolerance with our many discussions at late night and their friendship to look after me.

I also want to thank my classmates, Jen-Chien, I-Jeh, Min-Tsong, Chia-Hung, Meng-Lun, Hung-Chun, Shyh-Shun, Min-Yee, Yi-Hui , Wei-Shun, Po-Shuen, Bor-Shyh for their backing me up and taking care of me.

摘要

此論文提出一個人類健康長壽及其風險的免疫發炎模型。發炎導致大多數疾病（癌症，動脈粥樣硬化，老化相關疾病），為疾病共同引發途徑和主要風險，並導致最多病痛和死亡。

此論文模型，強調用非化學的抗發炎措施預防和治療疾病的可行性：消除食物抗原，食物毒素；解除精神壓力；消除隱性感染；和維持一個有效率的先天免疫。我們通過非化學非侵入性的方法，使用電子生物反饋，作為一種對抗發炎的措施。電子生物反饋抗炎反射效率很高，兼顧非化學非侵入性，為此一做法在每個人日常生活用以預防疾病，加強了基礎。

論文提出可能大幅延長人類無病痛之壽命的理論。用目前已有的自體先天免疫 NK 細胞增殖技術改變免疫表現型（immune phenotype）成為 NK 細胞優勢表現型，可能可以延後人類疾病，癌症，動脈粥樣硬化，老化發生, 減少病痛和死亡，大幅延長可能壽命。

先天免疫細胞治療癌症延長人類壽命的案例研究證明，利用自己的先天免疫治療癌症是可行的，合理的。四個末期癌症案例研究中。一末期肝癌患者，以自體先天免疫細胞 NK 細胞輸血作為唯一的治療。 肝癌及門靜脈血栓完全消失！自體 NK 細胞輸血延長所有末期癌症案例的生存，延長無症狀間隔，延長病程無惡化間隔。它可以與任何療法組合使用。病人壽命生活品質大幅優於其他未併用 NK 治療方式的末期癌症患者。

自體先天免疫細胞 NK 細胞的免疫表現型優勢地位，不僅有助於治療癌症是，也有助於預防動脈粥樣硬化。在冠心病豁免人口中，自然殺手細胞（NK）的 CD 56 暗淡子型（CD 56 Dim subtype）是優勢免疫表現型。相反的，冠心病患者免疫表型中的，自體先天免疫細胞 NK 細胞表現劣勢。NK 細胞是健康長壽的免疫表型。具 NK 細胞的優勢免疫表現型的老人有較佳內分泌樣貌，較多的肌肉，生活較活躍，較少感染發炎，較少病痛，較長壽。

NK 細胞治療和預防癌症。NK 細胞可以防止動脈粥樣硬化。NK 細胞協助治療各種感染，使發炎重回到平靜。NK 細胞的優勢表型的是健康老齡化。靜脈周血有容易取得到豐富的 NK 細胞前體細胞，使先天免疫細胞 NK 細胞的優勢免疫表現型可以輕易達成。

非化學抗發炎，以及非化學自體先天免疫細胞 NK 細胞解決方案，將是一個非常重要的防止病痛和死亡的方法。每個人在日常生活中對解除免疫負荷，解除精神壓力，非化學抗炎措施，達成自體先天免疫細胞 NK 優勢表現型，將有效地降低癌症，動脈粥樣硬化，許多老化有關的疾病的發生。

善待免疫的生活方式，降低發炎，優先使用對免疫無害的有效方法治療疾病，並促進或改變成為先天免疫細胞 NK 優勢表現型，人類壽命將有可能大幅超越目前的限制。

關鍵字：長壽、疾病的發病機制、統一的理論、免疫、發炎、抗發炎反射、精神壓力、心率變異性、非化學、免疫療法、細胞療法、自然殺手細胞、腫瘤、動脈粥樣硬化、老化、NK 細胞優勢、免疫表現型

ABSTRACT

In this thesis we propose an immune –inflammatory model of human longevity and risks. This thesis will theorize that inflammation as a common pathway and major risk leading to most diseases (cancer, atherosclerosis, aging-related diseases).and leading to most morbidity and mortality. Anti-inflammatory measures are going to slow or prevent diseases from major health risk, inflammation. And the thesis will also theorize immune phenotype change by available cell proliferation technology would extend current limitation of human longevity.

There are many non-chemical approaches in disease prevention and treatments. Of these, electrical bio-feedback is an efficient non-invasive non-chemical approach against inflammation. The nature of non-invasiveness and the efficiency of this approach strengthens the basis for using electrical bio-feedback anti-inflammatory reflex in disease prevention for everyone in daily life. Here, the result showed that HRV-HR is a promising parameter for checking inflammation. Other non-chemical approach are also emphasized in disease prevention and treatment: Eliminate of food antigen, food toxins; Unload mental stress; Eradicate occult infection. And maintain an efficient innate immunity.

This thesis also showed that changing immune phenotype is available by cell proliferation technology. The case study on cancer innate immune therapy proves that it is possible and reasonable to use our own innate immunity to treat cancer. Autologous natural killer(NK) cell transfusions eradicate cancer in pilot study. Terminal hepatocellular carcinoma with portal vein thrombosis totally disappeared with NK cell transfusion as sole treatment. NK cell transfusion prolongs survival in all cases. NK cell transfusion lengthens symptom-free intervals and progression free intervals. It is freely used in any combination therapy. The life quality of patient is much superior

to otherwise treated patients. The phenotype change toward NK cell dominance is not only useful in treating cancer is is also useful in prevention against atherosclerosis.

The phenotype of NK cell, CD 56 dim played an important role in Coronary Heart Disease. This type of patients has less NK cells. NK is the immune phenotype of healthy longevity. Those with higher NK has favorable endocrine profile, more muscle, more life activity, and less morbidity. Moreover, NK treats and prevents cancer, atherosclerosis, treats variety of infection and brings inflammation back to homeostasis. Abundant NK precursor cells are easily available in the peripheral blood.

NK treats and prevents cancer. NK prevents atherosclerosis. NK cell treats variety of infection and brings inflammation back to homeostasis. NK prevalent is the phenotype of healthy ageing. There is easily available abundant NK precursor cells in the peripheral blood , that make change into NK phenotype type present and practical by current proliferation or promotion technology.

. Thus, we conclude that by having your life immune-friendly way, by lowering the inflammation to prevent diseases, by selecting effective and immune-harmless treatment against diseases, and by promoting or changing into NK prevalent immune phenotype, would be the key for the human being to live beyond the current limitation of human longevity.

Keyword: Longevity, diseases pathogenesis, unified theory, immune, inflammation, anti-inflammatory reflex, mental stress, heart rate variability, non-chemical, immune therapy, cell therapy, natural killer cell, cancer, atherosclerosis, ageing, NK prevalent , immune phenotype.

TABLE OF CONTENTS Page

LIST OF FIGURES

CHAPTER 1 INTRODUCTION

1.1 Background

Cancer, coronary heart disease, cerebral vascular disease, diabetes, liver diseases like hepatitis, cirrhosis, kidney diseases like glomerulonephritis, chronic renal failure, are at present the most leading etiology of morbidity and mortality. For people to live longer and healthier, we must devote more effort to understand these diseases, to search in depth the relevant risks, and engage to fight against these diseases.

Current approaches of diseases diagnosis, treatments and prevention have their limitations: First, it lacks of integration, tends to know superficial facts, signals, signs, symptoms, diseases, without knowing the common causative problems. Secondly, it tends to have "Head to head, feet to feet" approach of treatments. This often leads to mutual conflicts, harmful treatments, and unsuccessful treatments. Last but not the least, it lacks central concept of disease pathogenesis and prevention. It is very difficult for common population or even clinicians to have an operable model of diseases prevention.

In this thesis we proposed a model: **An Immune-Inflammatory Model of Human Longevity and Risks**. (Figure 1) This thesis will theorize that inflammation is the common pathway and major risk leading to most diseases (cancer, atherosclerosis, aging-related diseases).and leads to most morbidity and mortality. Anti-inflammatory measures are going to slow or prevent disease progression. We also conclude that immune phenotype change by available cell proliferation technology would extend current limitation of human longevity.

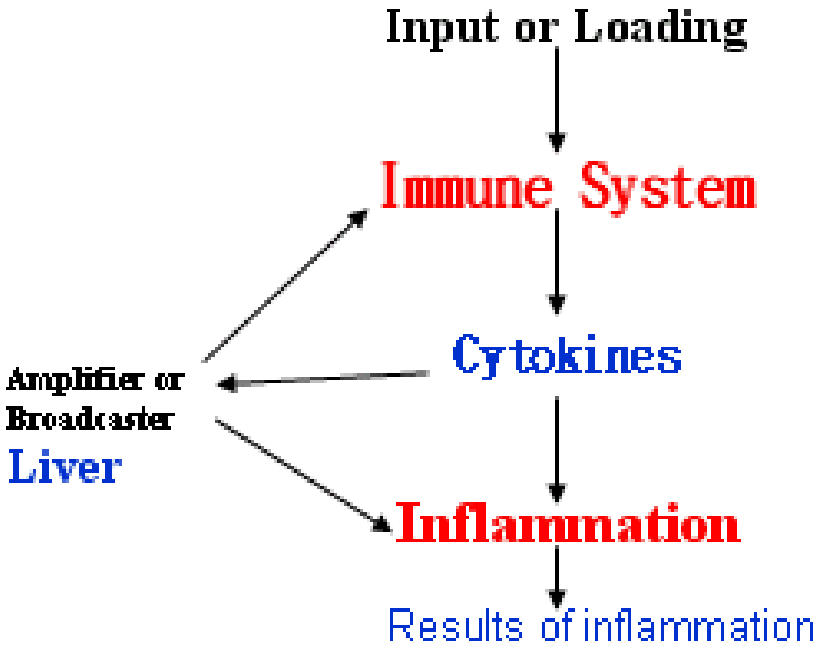


Figure 1 Block Diagram of the Model

Figure 1. Block Diagram of Six candidates of the Immune-Inflammatory Model of Human Longevity and Risks

1.2 Motivation , Objective and Three Questions to be Answered

There are three questions to be answered.

Is it possible to treat diseases, like cancer, through the inner power of our own?

What is the extent that immunity can help ?

Is it possible to prevent diseases and achieve longevity through the power of our own?

The motivations of this thesis were to response to the above problems and provide the following issues ; 1. To integrate disease pathogenesis and discrete phenomenon into one unified theory. 2. To develop theory basis for non-chemical anti-inflammation measures for every one to prevent diseases in daily life. 3. To eliminate current conflict, harmful, unsuccessful treatments. 4. To facilitate the care quality of cancer patient. 5. To extend current limitation of human longevity.

In this thesis we hope to conceptualize that inflammation is a very important pathway of disease pathogenesis and a major obstacles to longevity. We can also test the efficiency of non-chemical bio-feedback anti-inflammation measures. We also hope to theorize the innate immunity NK cell proliferation technology as a feasible one for human to extend current limitation of longevity. Through these, the basis of our bio-signal profile platform of immune system and related others (eg. HRV-HR) could be build.

1.3 Our Previous Work

To meet theses goals, some related biomedical signals or medical informatics investigations have been done and published by the author or by our lab colleagues. These works include the innate and adaptive Immune cell profile by flow cytometry and the beta-glucan stimulated NK/lymphocyte profile [15], the study biosignal platform; Portable device for ECG[23], Study on heart rate variability [24], Automatic detection of FMD(Flow Mediated Dilatation), into study of immune, inflammation, atherosclerosis, HRV[21,22], Security transmission of biomedical signals[19] and the study of biomedical informatics platform[20].

1.4 Some Progress of Immune Based Cancer Therapy: Cell therapy

Treatment for advanced cancer is usually very torturous. Main treatments of cancer include surgery, chemotherapy, radiation therapy. Surgery treatment removes identifiable cancer and the surrounding tissue with related complications, bleeding, infection, and anesthesia risks. Using chemotherapy to treat cancer causes a lot of side effects. Besides killing the cancer cells, it may alter the immunity and damage the body. Radiation therapy against cancer has side effect of radiation damage and has a lot of

limitations, for example dosage limit, responsiveness. Chemotherapy and radiation therapy has also bone marrow suppression side effect, leading to anemia, poor immunity, bleeding tendency.

Here in Taiwan and China, I have been taking care of patients using autologous cell therapy to treat the most notorious disease, cancer. The patients receive autologous NK cell (Natural Killer cell) of innate immunity under a government-approved clinical trial. Several miracle cases, originally regarded as terminal cancer, survive much longer than ever expected. Some of them even have their cancer totally disappeared. The hospitalization days were minimal or even none because the course is smooth and uneventful. Moreover, all the common symptoms of cancer, namely cachexia, pain, nausea, weakness, fever, seldom or never occurred.

Autologous cell therapy is a means to use cells from our own to treat diseases. It is a less-chemical approach to treat disease with less or even no medications, through the power of our own.

Contrary to other main treatment, using NK cell to treat cancer has no complication related to surgery. No torturous side effect of chemotherapy. No tissue damage, no bone marrow suppression. Patients tolerate the treatment very well. Almost all patients feel better. Autologous cell therapy could be freely combined with any other cancer therapy.

1.5 Dissertation Organization

In this dissertation, the background, the block diagram, the motivations and objectives of the thesis, previous works and recent advance of immune cell therapy are described in Chapter 1. The rest are organized as following.

Chapter 2 describes the unified immune-inflammatory theory model of

longevity and diseases; the six candidates of the model: input, immune system phenotype, cytokines, broadcaster and amplifier, the inflammation, the results of the inflammation; inflammation as major risks; diseases, cancer, atherosclerosis, ageing as the results of inflammation of results; and how to use this model to prevent diseases and achieve longevity.

Chapter 3 describes inflammation case study: the relationship of mental stress and inflammation; indexed with heart rate variability and heart as surrogate marker of autonomic system and inflammation interaction; the application of non-chemical biofeedback as an effective measure to lower inflammation.

Chapter 4 describes the NK of innate immunity as a longevity phenotype against cancer, ageing, atherosclerosis; NK identification and subtyping flow cytometry platform; why NK is promising through our current technology.

Chapter 5 describes NK case study; NK transfusion as an safe and applicable phase 3 clinical technology; the promising preliminary results in four previously regarded as terminal cancer patients.

Chapter 6 concludes the thesis.

CHAPTER 2 AN IMMUNE-INFLAMMATORY MODEL OF HUMAN LONGEVITY AND RISKS

2.1 A Few Words Ahead the Model

There is an old saying: **We are what we eat.**

There is pretty much truth in it. Our body has been developed from one cell to what we are today. The composition of individual body equates to input minus output. External or environmental inputs interplay with our genotype to become our phenotype, that is, what we are.

The Immune system is our first front line to respond to external inputs; food, air, water, microorganisms, various kinds of energy, or injury. When the immune system is challenged by infection, antigen, toxin, physical injury, and mental stress, that may actually damaged the cell or may regarded as threat to the cell , it will evoke a cell injury signal, the inflammatory signal. The immune system has to solve the problems itself and would arouse inflammatory response from the immune system.

2..2 The Immune- Inflammatory Model

As shown in figure 2, this model is makeup from six candidates. Candidate 1 is the Input, or loading into immune system. Candidate 2 is the response network, immune system phenotype. Candidate 3 is the communication signal, Cytokines. Candidate 4 is the Amplifier and broadcaster. The liver will release acute reactive protein into circulation of the body. Candidate 5 is the Landmark events: Inflammation. The last candidate is the Results of inflammation. It consists of biomedical signal, clinical markers, signs, symptom, and type of diseases. This module linked together all seemed to be independent thread of events. This will provide chance to solve the puzzle the

right way, the immune way.

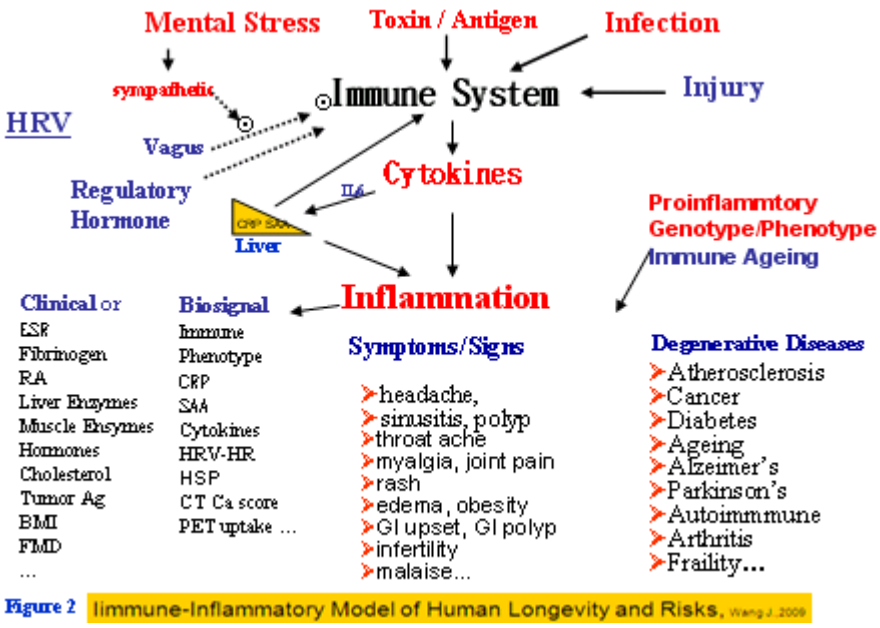


Figure 2: The immune-inflammatory model of human longevity and risks. The names of these symbols are as follow: ESR erythrocyte sedimentation rate; RA rheumatoid antigen; Tumor Ag tumor antigen; BMI body mass index; FMD flow mediated dilatation; CRP C-Reactive Protein; SAA serum amyloid antigen; HRV heart rate variability; HR heart rate; HSP heat shock protein; CT computer tomography; PET positron emission tomography; GI gastrointestinal

2.3. Immune Loading:

Loading to the Immune system is rather complicated, it consists of both internal and externals events.

2.3.1 Antigen or Toxin

External antigen, self antigen(eg.HSP), and toxin(smoking, drinking, oxidized-LDL) will induce inflammation

2.3.2 Infection

Bacteria, virus, fungus, or unresolved previous infection will load the immune system. Long term effect of chronic loading compromises the capacity of immunity.

2.3.3 Mental Stress

Generally, people less often addressed as immune loading is internal input from mental stress. Through vagus nerve, the brain has an anti-inflammatory reflex. Uncompensated repeated mental stress overall causes immune loading and inflammation.

There's an anti-inflammatory reflex of autonomic nervous system via para-sympathetic vagus nerve. Increase heart rate and decrease Heart rate variability (SDNN, standard deviation of normal to normal R wave) is correlated with CRP elevation and inflammation. (Lombardi, 2004 , [9) This anti-inflammatory reflex mechanism links mental stress to immune loading, inflammation, and the relationship with many diseases.

2.4. The Inflammatory Response and the Amplifier

When the immune network is triggered by the input or loading, cytokines will be released, and inflammatory response will be initiated. Inflammatory response is composed of both local events and a systemic activation. When systemic activation, the **liver** will serve as amplifier, amplifying the biosignal, release acute reactive protein, CRP, SAA broadcasting via circulation.

The liver is mainly built to broadcast the immune loading from what we eat. Food is the largest input to our body, bacteria, virus, fungus, food toxins, water toxins could enter our body in largest amount compared to other entry. That is why our

gut –associated lymphoid tissue comprise 90% of lymphoid tissue. The gut-guarding liver will respond to other sources of blood circulating cytokines as well. Its huge capacity of synthesis could scale-up amplify the inflammation in a reentry circle.

2.5. The Results of the Inflammation.

Inflammatory results composed of molecular events, cellular events and systemic events. Inflammation is a network of the interactions of these events.

2.5.1 Symptom/Sign

The events could be perceived as symptom. Without the knowledge of what is going. The person will neglect it which might leads further deterioration. A lot of clinical symptom sign originally interpreted as unrelated events and the physician or patient used to treat it" head to head, foot to foot'. It is wise to interpret ate again in this immune– inflammatory model for us to understand the pathogenesis and take action to do the right things to prevent from further degeneration into more serious one.

2.5.2 Biosignal / Clinical Markers

The markers of the interaction include WBC, IL-1 IL-6, TNF, INF, CRP, SAA etc,. A lot Clinical markers related to inflammation, mention a few first, ESR, fibrinogen, coagulation profile, PAI-1, RA, ferritin, liver enzyme, muscle enzyme, tumor antigen, steroid hormone profile, HR, HRV, MDCT calcium score, PET uptake etc.

It is very useful to detect, to intepretate, to monitor, to intervene this inflammatory or anti-inflammatory markers.

2.5.3 Diseases from Inflammation

If tissue health is not restored or with stable low grade irritation, inflammation becomes chronic condition that continuously damages the tissues . During chronic inflammation, tissue injury and healing proceed simultaneously. The damage caused by chronic inflammation accumulates slowly, sometimes asymptomatically for years, may eventually lead to severe deterioration

Before 1800, adult life expectancy was only about 40 to 50 years. Today, the immune system must remain active for much longer. This very long activity may leads to chronic inflammation that slowly damages the organs. This a typical phenomenon linked chronic inflammation to ageing and is considered the major risk factor for age-related chronic diseases. Alzheimer's disease, atherosclerosis, diabetes , cancer – have an important inflammatory component, Liver cirrhosis, hepatitis, viral , alcoholic, autoimmune have important inflammatory component. So does chronic renal failure associated with glomerulonephritis or glomerulonephropathy. And major lung diseases, cancer, pneumonia, COPD as well.

Our immune system has evolutionarily programmed to control pathogens, so pro-inflammatory responses are likely to resist fatal infections aggressively. Thus, inflammatory genotypes are a necessary part of the normal host responses to pathogens in early life, but the overproduction of inflammatory molecules might cause inflammatory -related diseases and eventually death later. Therefore, low responder genotypes involved in regulation of innate defense mechanisms, might better control inflammatory responses and age-related disease development, resulting in an increased chance of long life survival in a "permissive" environment with reduced pathogen load, medical care and increased quality of life. (Licastro et al. 2005,

[8])

2.5.4.Ageing from Inflammation

"Inflammaging" has been coined to explain the underlining inflammatory changes common to most age-associated diseases.. This new term means that ageing is coupled with an age- dependent up-regulation of the inflammatory response, due to the chronic antigenic load which triggers the onset of inflammatory disease. Inflammaging , the up-regulation of a variety of anti-stress responses at the cellular and molecular level, is the consequence of the body's ability to counteract and modulate the effects of a variety of stressors, which cause the accumulation of molecular and cellular scars(Francechi et al. , 2000 ; Francechi et al, 2003) Cytokine production may also be up-regulated 'in vivo' in old subjects resulting in an abnormal elevation of proinflammatory cytokines during inflammatory responses (Licastro et al., 2005, [8]).

Evidence suggests that pro-inflammatory genotypes are related to unsuccessful ageing, and, contrastly, controlling inflammatory status may allow a better chance of successful ageing. So, age-related diseases are "the price we pay" for a life-long active immune system.

Aged macrophages also contribute to an impaired the proliferative response of activated peripheral T lymphocytes (Pawelec et al,1998 , [11]).

Aged phagocytes, such as macrophages showed an impaired respiratory burst and reactive nitrogen intermediate production with a decreased ability to destroy pathogens; Aged dendritic cells were less efficient in activating both T and B cell populations (Plackett et al. , 2004, [14]).

The 'in vitro' production of pro-inflammatory cytokines, such as IL-1, IL-6

and TNF-α, by mitogen activated peripheral blood monuclear cells from elderly persons was higher than that from young donors (Licastro et al., 2005, [8])

2.6 How May Use this Model to Prevent Diseases and Extend Longevity

In the clinic, diseases, symptoms/signs, biomarker, biosignal all has immune-inflammation relevance. Integrate the discrete findings, treat the causative problem if possible. Specific immunity against pathogens and mutated cell should be always preserved when we select the treatments. It will largely avoid conflict and harmful treatments.

In general life, non-chemical approach are emphasized in disease prevention and treatment. First of all, Unload your immune system: Eliminate of food antigen, food toxins;. Unload mental stress; :, Eradicate occult infection. And maintain an efficient innate immunity. Second, control over reactive inflammation by anti-inflammatory reflex, physical therapy, and functional food. Third, we should have regularly follow up.to monitor inflammation/ diseases/immunity. Then select immune –friendly treatment to control inflammation and diseases.

The following figure(Figure3) outline the use of the immune-inflammatory model to prevent diseases and prolong longevity.

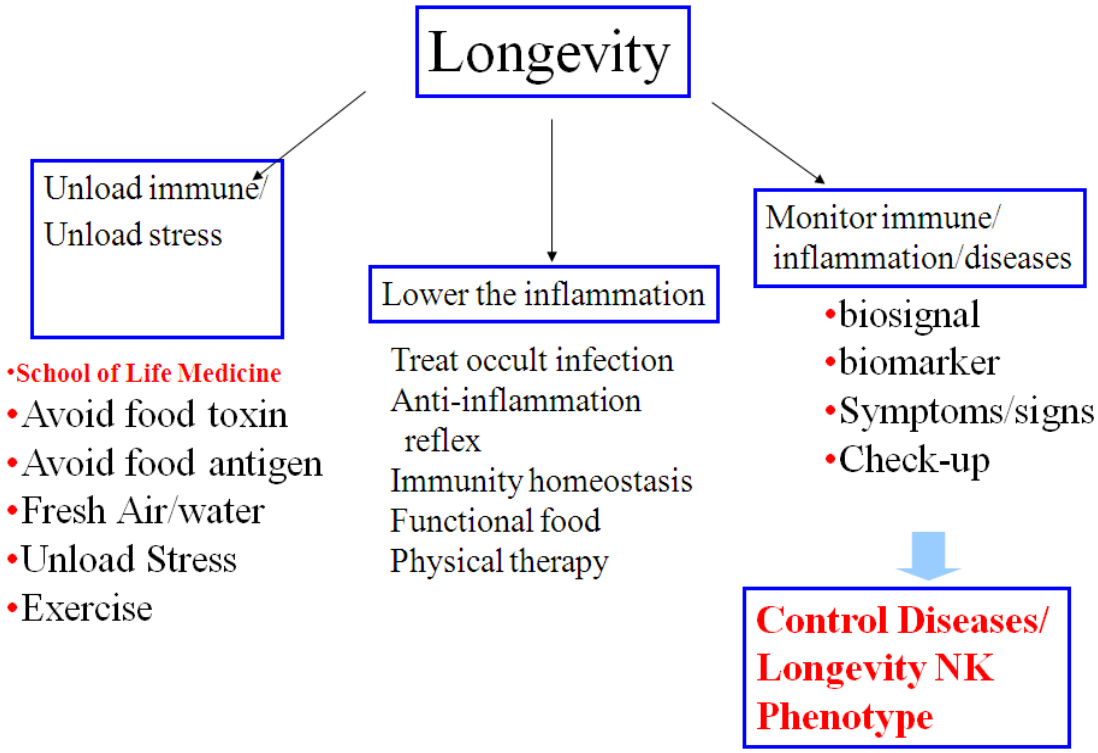


Figure 3 stepwise use of immune-inflammatory model to control disease and extend longevity

Chapter 3 INFLAMMATION CASE STUDY

3.1 Inflammation Associated with Reduced Heart Rate variability(HRV)

Besides general available biomarker, like hs CRP, as useful tool to detect subclinical inflammation. There is a an approach through heart rate variability. The study on heart rate availability in middle-aged and elderly Inflammation is associated with reduced heart rate availability (Sajadieh et al., 2004 ,[12]).

Heart rate and SDNN has been found negatively associated with inflammation markers. In multivariate analysis both increase heart rate and reduced heart-rate variability were associated with sub-clinical inflammation. This study is of relevant clinical interest.The negative association between increased heart rate and reduced HRV and C-reactive protein indicates increase sympathetic tone characterized patients with of sub-clinical inflammation. Both sympathetic activation and subclinical inflammation have been independently associated with an increased cardiac mortality. The combination of heart rate , HRV and CRP could be used to detect health risk. And might reflect an interaction between the autonomic nervous system and inflammation, autonomic imbalance leading to sympathetic predominance may promote activation of inflammation, which, in turn, could further alter autonomic balance. This could explain the presence of signs of sub-clinical inflammation in clinical conditions such as diabetes, hypertension and coronary artery disease with incerased signs of sympathetic tone and decreased reduced vagal tone (Lombardi, 2004, [9]).

3.2 Anti-Inflammatory Reflex by Electrical Biofeedback(Vagal Stimulation)

The anti-inflammatory Vagal stimulation is effective to save life even in the setting of most severe infection, sepsis.

We used electrical biofeedback device to study heart rate variability and heart rate.The biofeedback device is an ultralow frequency electrical stimulator. It is a health device recognized by Japan and other countries include ROC. It is used to stimulate ANS via electrical stimulation. We try using it as a non-chemical approach hypothesis to lower inflammation. The test was conducted on approved effect of shoulder pain, joint pain, insomnia via anti-inflammatory reflex. A total of 60 volunteers age between 35 to 76 with baseline normal sinus rhythm took part in this experiment.

3.3 Experimental Design:

Before testing, the subject is kept sitting at the device for at least 5 minutes. Then we record BP and resting ECG signal for 5 minutes. The electrical stimulator is in operation for 30 minutes. We repeat obtaining BP and resting ECG. Later the ECG is under analyzed through the following protocol.

3.4 Signal Processing

The signal process is shown in figure 4. The ECG signal is first differentiate (Figure 5). After performing Hilbert transform on the differential signal, Peak detection method is used. The detected peak is the R-wave on the ECG (Figure 6). Then this R-wave will be under review by a cardiologist to mark ectopic beats. Then the duration time between the two R-wave is calculated to find RR Interval. Baseline heart rate was calculated by mean RR interval and heart rate variability was calculated as standard

deviation of normal to normal RR interval(SDNN).

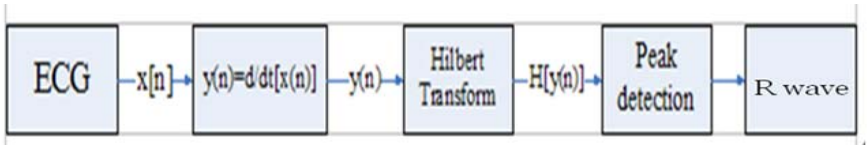


Figure 4 processing of ECG to identify R wave

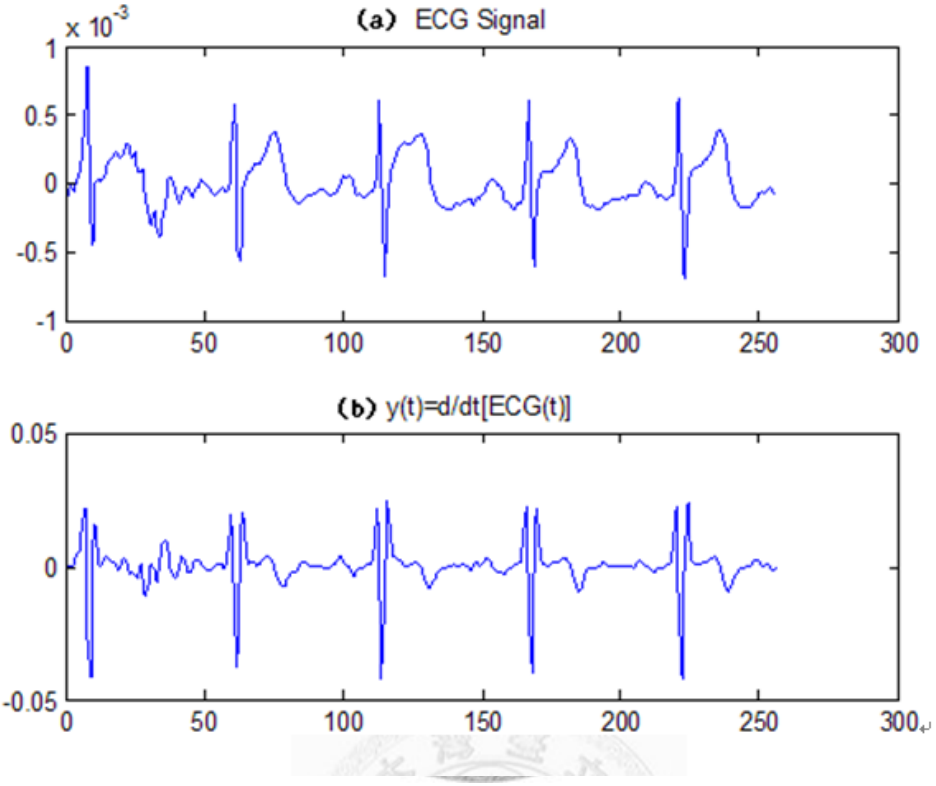


Figure 5 (a)ECG (b)ECG differential

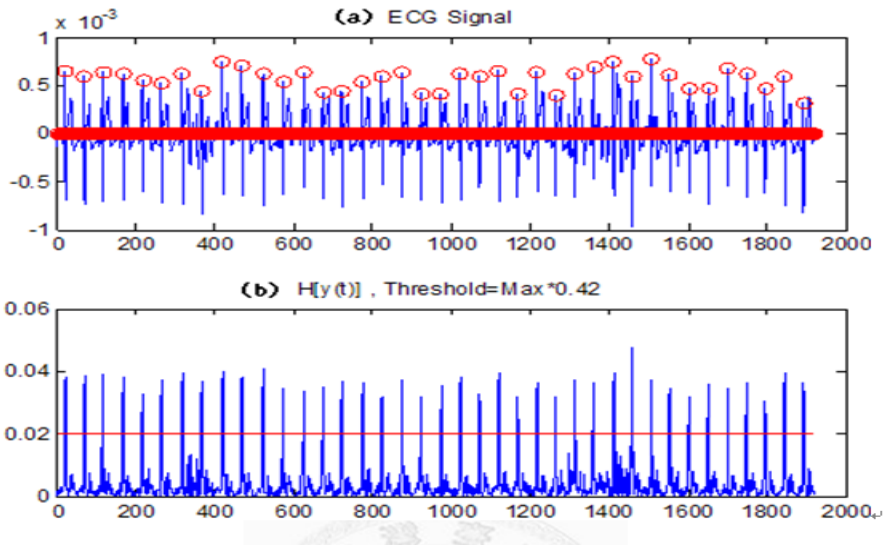


Figure 6 processing of ECG to identify R wave

3.5 Results and Conclusion

The HRV-HR result is shown in figure7. The plot of SDNN and HRV lowering is shown in figure 8.

Favorable response is defined as both increased SDNN and decreased HR;

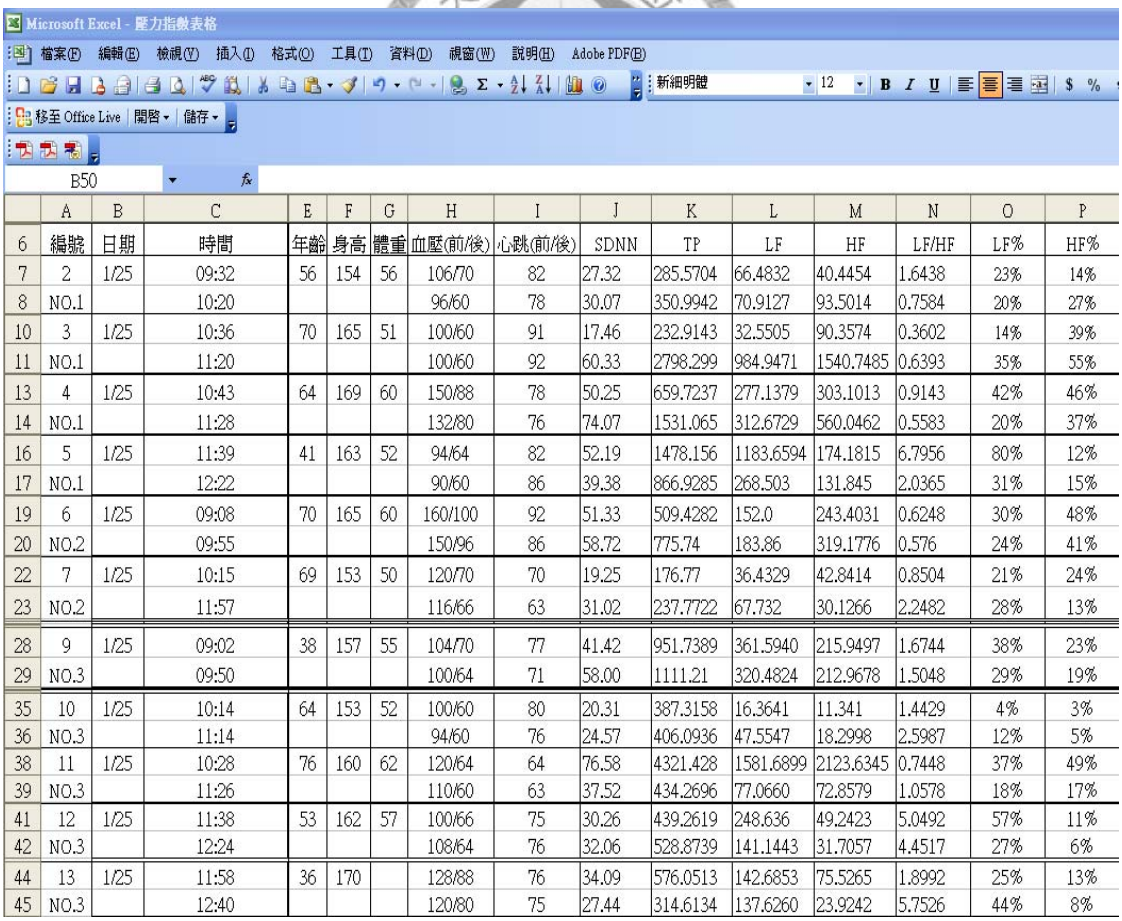
Unfavorable response is defined as decresed SDNN and increased HR

Results: 61.7% (37/60) subjects shows favorable response(after 30mins stimulation;

10% (6/60) subjects shows unfavorable response;

28.3%(17/60)subjects show equivocal response

For this biofeedback device in term of typical anti-inflammatory reflex there is 61.7% responder. Thus this device is a acceptable supplement for anti-inflammatory.



Microsoft Excel - 壓力指數表格

檔案(F) 編輯(E) 檢視(V) 插入(I) 格式(O) 工具(T) 資料(D) 視窗(W) 說明(H) Adobe PDF(B)

新細明體 12

移至 Office Live | 開啓 | 儲存

B50

	A	B	C	E	F	G	H	I	J	K	L	M	N	O	P
6	編號	日期	時間	年齡	身高	體重	血壓(前/後)	心跳(前/後)	SDNN	TP	LF	HF	LF/HF	LF%	HF%
7	2	1/25	09:32	56	154	56	106/70	82	27.32	285.5704	66.4832	40.4454	1.6438	23%	14%
8	NO.1		10:20				96/60	78	30.07	350.9942	70.9127	93.5014	0.7584	20%	27%
10	3	1/25	10:36	70	165	51	100/60	91	17.46	232.9143	32.5505	90.3574	0.3602	14%	39%
11	NO.1		11:20				100/60	92	60.33	2798.299	984.9471	1540.7485	0.6393	35%	55%
13	4	1/25	10:43	64	169	60	150/88	78	50.25	659.7237	277.1379	303.1013	0.9143	42%	46%
14	NO.1		11:28				132/80	76	74.07	1531.065	312.6729	560.0462	0.5583	20%	37%
16	5	1/25	11:39	41	163	52	94/64	82	52.19	1478.156	1183.6594	174.1815	6.7956	80%	12%
17	NO.1		12:22				90/60	86	39.38	866.9285	268.503	131.845	2.0365	31%	15%
19	6	1/25	09:08	70	165	60	160/100	92	51.33	509.4282	152.0	243.4031	0.6248	30%	48%
20	NO.2		09:55				150/96	86	58.72	775.74	183.86	319.1776	0.576	24%	41%
22	7	1/25	10:15	69	153	50	120/70	70	19.25	176.77	36.4329	42.8414	0.8504	21%	24%
23	NO.2		11:57				116/66	63	31.02	237.7722	67.732	30.1266	2.2482	28%	13%
28	9	1/25	09:02	38	157	55	104/70	77	41.42	951.7389	361.5940	215.9497	1.6744	38%	23%
29	NO.3		09:50				100/64	71	58.00	1111.21	320.4824	212.9678	1.5048	29%	19%
35	10	1/25	10:14	64	153	52	100/60	80	20.31	387.3158	16.3641	11.341	1.4429	4%	3%
36	NO.3		11:14				94/60	76	24.57	406.0936	47.5547	18.2998	2.5987	12%	5%
38	11	1/25	10:28	76	160	62	120/64	64	76.58	4321.428	1581.6899	2123.6345	0.7448	37%	49%
39	NO.3		11:26				110/60	63	37.52	434.2696	77.0660	72.8579	1.0578	18%	17%
41	12	1/25	11:38	53	162	57	100/66	75	30.26	439.2619	248.636	49.2423	5.0492	57%	11%
42	NO.3		12:24				108/64	76	32.06	528.8739	141.1443	31.7057	4.4517	27%	6%
44	13	1/25	11:58	36	170		128/88	76	34.09	576.0513	142.6853	75.5265	1.8992	25%	13%
45	NO.3		12:40				120/80	75	27.44	314.6134	137.6260	23.9242	5.7526	44%	8%

Figure 7 HRV HR results

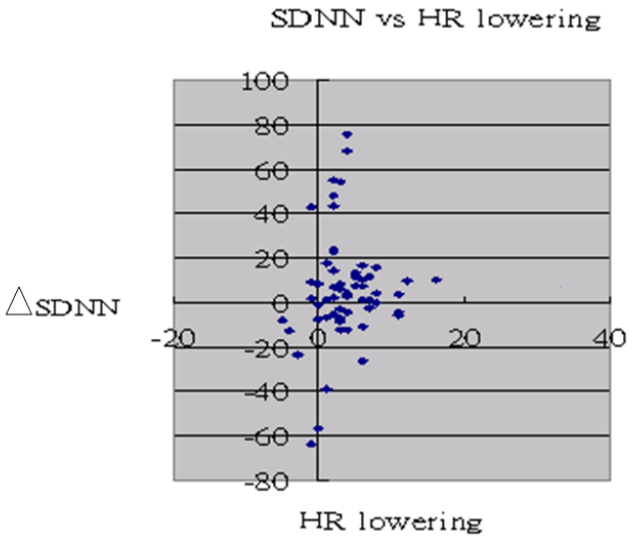


Figure 8 Plot of SDNN and HR lowering;

Chapter 4 NK CELL AS THEORY IDEA OF LONGEVITY PHENOTYPE OF LONGEVITY AGAINST CANCER, AGEING, AND ATHEROSCLEROSIS

4.1 NK Cell of Innate Immunity

The immune system may be classified into Innate(monocytes, natural killer and dendritic cells), and adaptive immunity (B and T lymphocytes). NK cells constitute up to 15% of peripheral blood mononuclear cells (PBMC) and are also found in peripheral tissues including lymph node, peritoneum, placenta and liver. The NK cell phenotype is characterized by lack of expression of the CD3 complex together with variable expression of CD16, CD56 and CD57.

Figure 9 depicts the process of identifying NK cell from lymphocyte cluster using mAb against CD3 , CD16 and its subset through different CD 56 level.

NK cell CD56bright and CD56dim subsets are defined basing on the intensity of CD56 expression. These subsets have different tissue homing properties due to different patterns of expression of cytokines receptors and adhesion molecules. The major CD56dim NK cells have a granular phenotype and exhibits more cytotoxic activity than CD56bright NK cells. CD56bright NK cells express low levels of CD16 and constitute less than 10% of circulating NK cells but are the dominant NK cell in lymph nodes.. CD56bright NK cells are the primary source of immunoregulatory cytokines such as IFN-γ, IL-10, IL-13 and GM-CSF.

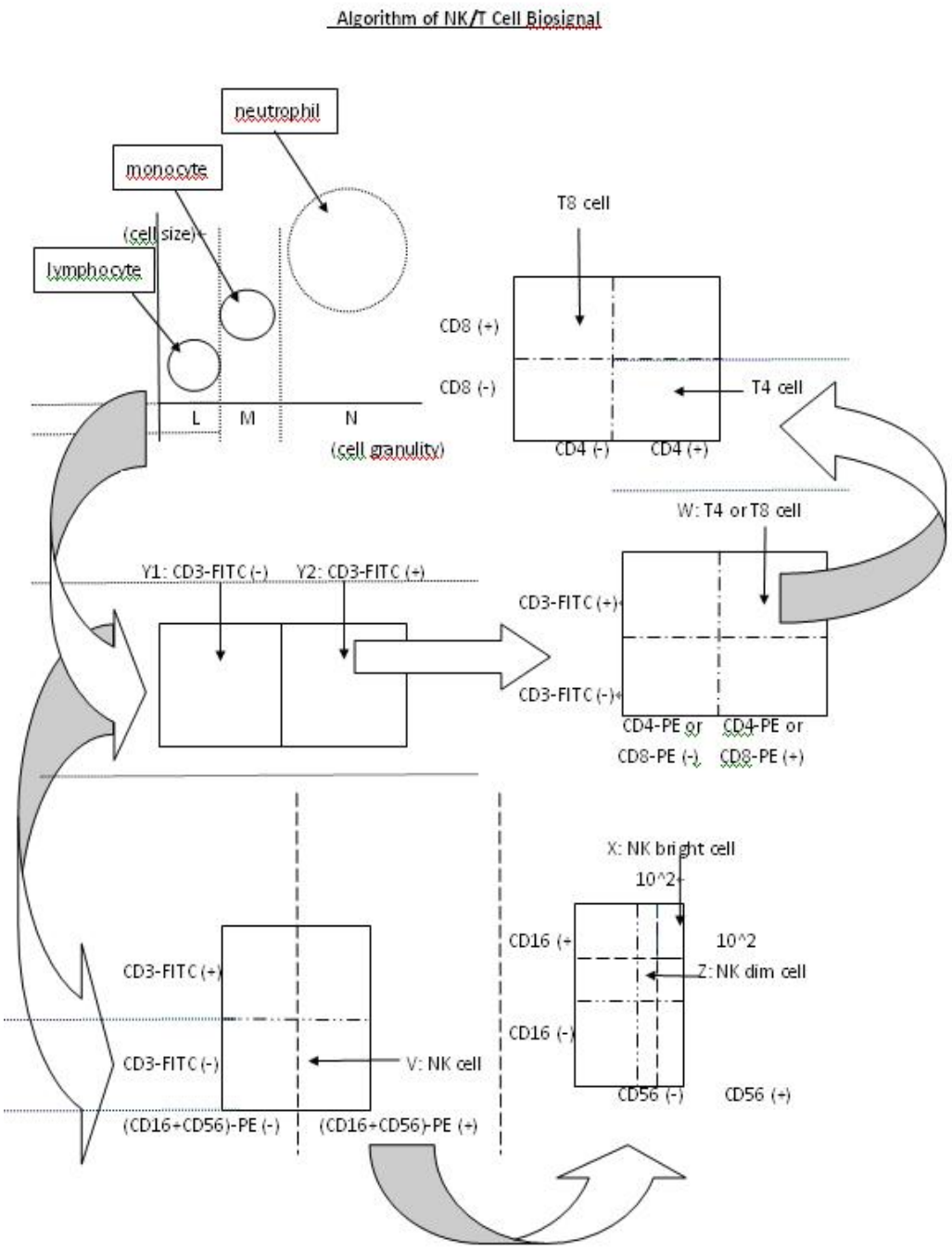


Figure 9 depicts the process of identifying NK cell from lymphocyte cluster using mAb against CD3 , CD16 CD56 and its subset through different l CD56 level.

4.2 NK Cell against Cancer

Natural killer (NK) cells were named for they defense against virus-infected and neoplastic cells in the absence of prior antigenic stimulation .

NK cells were discovered because of their ability to kill cancer cells in vitro. Depletion of NK cells in vivo facilitate tumor formation in several tumor models.(Hanna et al. 1985, [16], Wiltrout 1985[17]) Clear involvement of NK cells in anti-tumour immunity in vivo was shown in 1986 by Klas Kärre[6] with Major histocompatibility complex (MHC) class I in NK cell recognition. NK cells kill certain tumor cell lines in vitro even without significant levels of MHC class I on tumor. cell surface.

It has been clealy illustrated that The role of NK cell together with cell from adaptive immunity weights toward immunity aginst tumor. It is during the elimination phase, nascent tumor cells are destroyed by NK cells. (Bhardwaj , 2007[1])

In Taiwan, autologous NK cells were approved as the core trial treatment under a phase 3 clinical trial against liver cancer and lung cancer .

Figure 10 showed that when NK cell dominate, the trend is toward tumor immunity and suppress tumor growth. However when proinflammatory cytokines is the majority, the trend which is toward tumor growth.

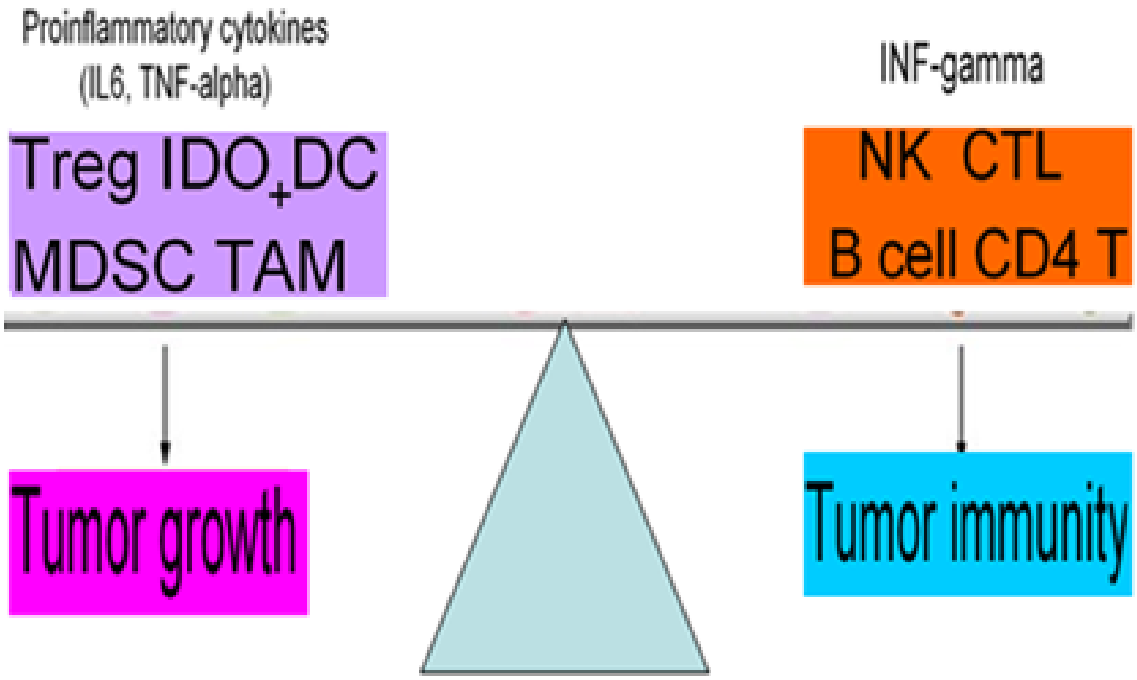


Figure10 Balance pivot for Tumor

4.3 NK Cell against Ageing

Ageing is associated with a decline in peripheral blood, CD56bright NK cells (Shivani et al., 2006[18]). NK activity in elderly subjects proved to be of predictive value of less morbidity and less mortality (Levy et al., 1991[7]).An elevated NK activity is correlated with well-preserved endocrine functions and muscular mass (Mariani et al., 1999[10]).

4.4 NK Cell in Longevity and Exempt from Atherosclerosis

Is it possible to prevent disease and achieve longevity through the power of our own?

What is the extent that immunity can help ?

4.4.1 Longevity and Exempt from Atherosclerosis

Common pro-inflammatory genotype has been associated with atherosclerosis and an genotype of "good control of inflammation" genotype protect against atherosclerosis. The number of oldest old people(>85 year old) has risen up of about 20 times , mainly contributed by the marked reduction of cardiovascular diseases prevalence rate , related to less antigen loading, less chronic inflammation, after the improvement of public hygiene. (Licastro et al. 2005[8])

4.4.2 NK cell of Innate Immunity and Exempt from Atherosclerosis

Another important study on innate immunity phenotype showed that Coronary Heart Disease patients had lower NK cytotoxic activity, decreased in the absolute number and percentage of total NK cell and CD3-CD56dim cytotoxic NK subset. The tendency is toward lower percentage of the CD3-CD56bright regulatory NK subset and CD3-CD56+IFN-γ+ cells in CHD patients (Hak et al, 2007[5])

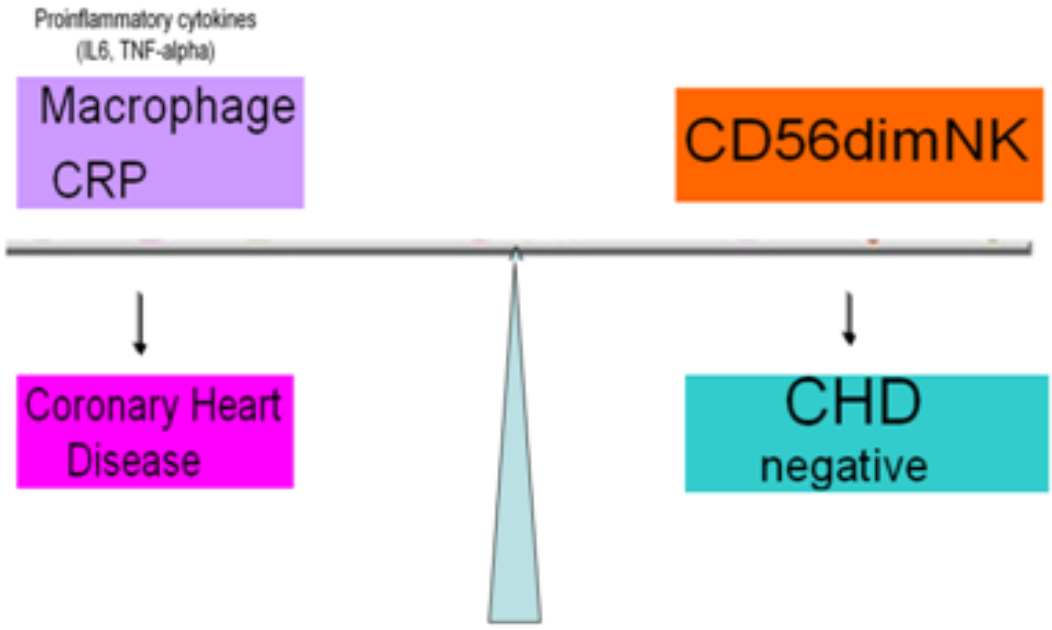


Figure 11 NK CD56dim dominance VS macrophage in coronary heart disease.

In Figure 11 we show NK CD56dim dominance is favorable immune phenotype exempt from coronary heart disease.

4.4.3 NK Cell as Longevity Phenotype

Our Hypothesis : A tentative Scenario for human to survive 85 year old or more the main determining factor would be exempt from cardiovascular diseases. The criterion is

High NK +/- High CD56dimNK +/- high CD56brightNK.

This immune profile would be a possible index for human longevity > 85 year old.

4.4.4 Why is NK?

The development of NK proliferation technology would be very useful. Contrast to stem cell with suspicious role in cancer stem cell. NK kills cancer; NK prevent atherosclerosis; NK is a accountable profile of longevity; NK precursor travel from bone marrow via blood to lymph node. It is easy obtainable the NK precursor from venous blood.

NK can back up exhausted adaptive T and B cell immunity without concern of marrow failure or thymus involution. Over 85 the major health risk is poor immunity. So for human to live longer and healthier. NK cell related approach deserve better attension.

Chaper 5 NK case study

5.1 Lymphocyte I mmunopheotyping by Flow Cytometry

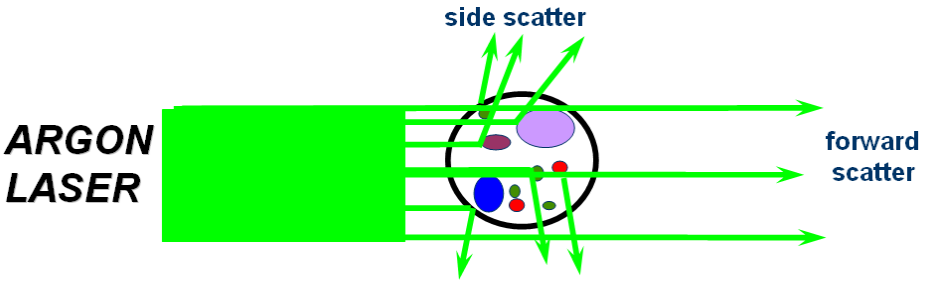


Figure 12 illustration of effect of size and granulity to forward scatter and side scatter

Using flow cytometry, forward scatter(FS) , side scatter (SS)pattern of laser could be used to identify the cluster of lymphoctye, the red dots in retangulated in the left side is the location of lymphocyte. middle, monocyte, right upper granulocyte.

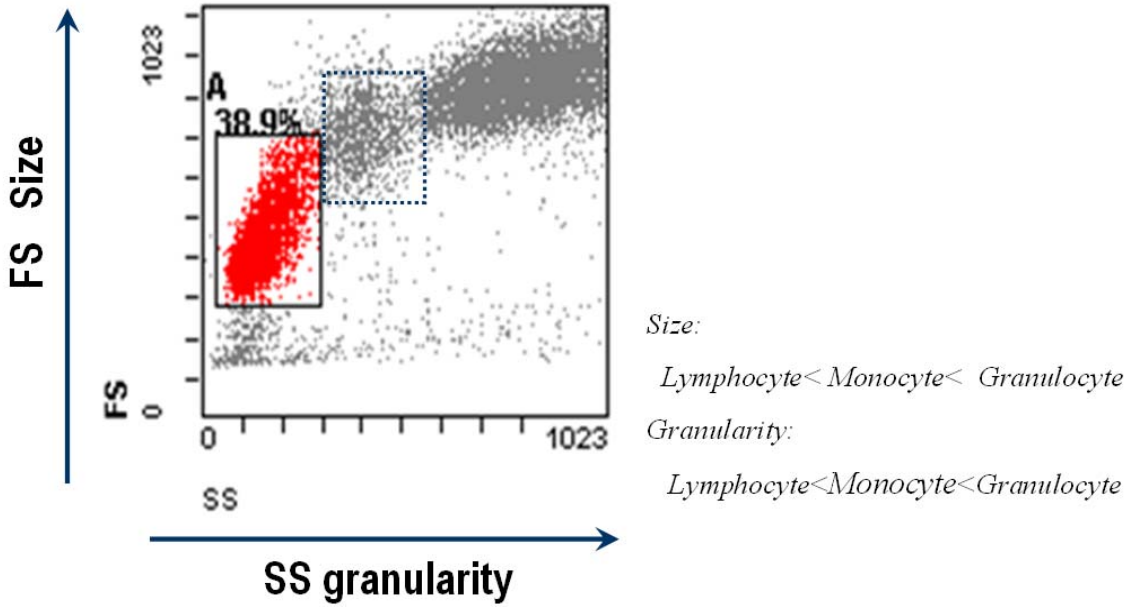


Figure13 distribution of lymphocyte by size(FS)and granulity(SS)

Lymphocyte Immunophenotyping

Antigen	Target cell recognized
CD3	Pan T cell
CD3+CD4	T helper cells
CD3+FOXP3	T reg
CD16+CD56	NK cells
CD19 or CD20	B cells
CD25	Activated cells
CD38	Activated cells
CD45	Pan leukocyte
CD45RO+CD4 or CD8	Memory T cells
CD45RA+CD62L+CD4 or CD8	Naïve T cells

Figure 14 1lymphocyte Surface antigen for phenotyping

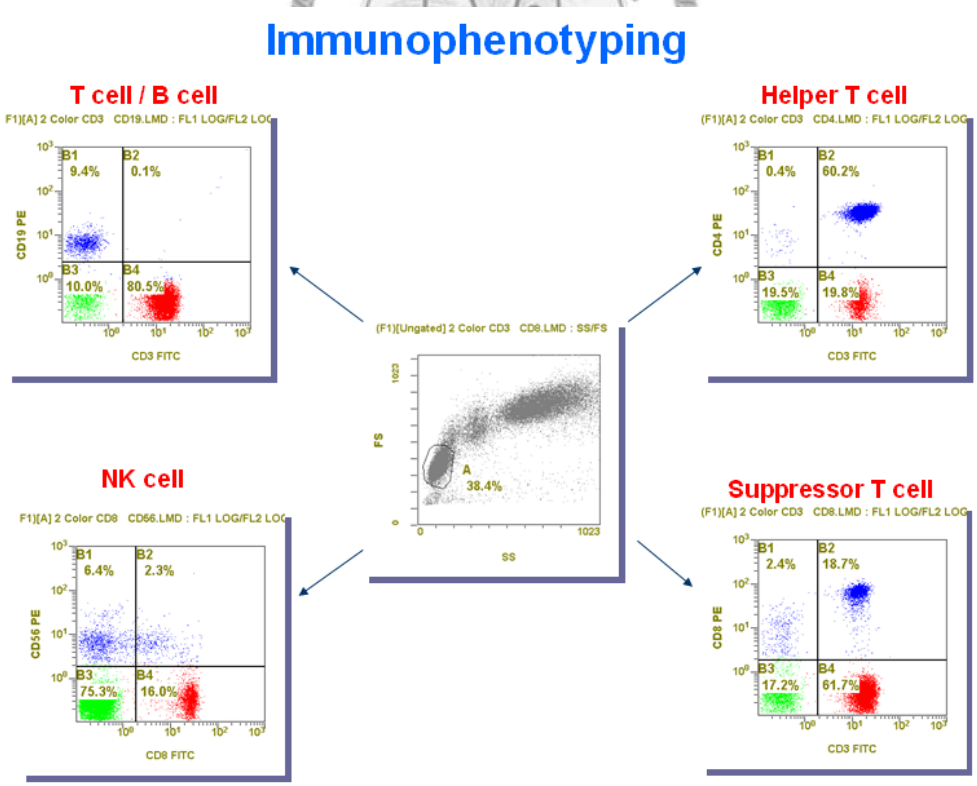


Figure 15 Uses MAb conjugated with fluorescent dye to classify the phenotype through SS FS signals in the gated lymphocyte area.

5.2 Fuctional Study and Cloning of NK Cell

Flow cytometry is used for fuctional study and cloning of NK cell Use more substype specific MAb, we can have NK phenotyping.

Natural killer cells are lymphocytes that lack CD3 and express CD16, CD56 and CD57. In recent years NK cells was categorized into two groups by level of CD56. Peripheral blood lymphocytes were stained with CD3, CD56 and CD16 . The CD3 negative lymphocyte was gated and ready to analyze CD56 and CD16 expression.In term of NK phenotyping, a easy expression is CD3-CD16+CD56+ lymphocyte. Stained with CD3 CD16 CD56 monoclonal antibody, the CD3 unstained lymphocyte was first gated, then gated with gate 1 and gate2 counting, results number of CD56Dim, CD56Bright.

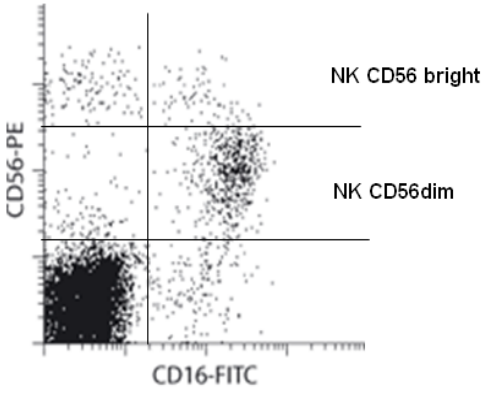


Figure 16 Flowcytometry results of NK and subtyping

As previously discussed, NK cell percentage, function, subtyping, are important in understanding disease pathogenesis, In the following section we will show our selected case study results.

5.3 NK Case Study 1 Hepatic Cancer Stage 4

Background

71 year old lady was presented to my clinic with diagnosis of hepatocellular carcinoma(HCC) stage 4.., She Bilateral liver lobes with multiple tumors, and portal vein thrombosis were noted. This old lady does not have habit of drinking, smoking. She has undelying chronic hepatitis with HBV, HCV, liver cirrhosis. No other major systemic diseases. Extremely poor prognosis was told by several medical centers. She refused chemotherapy suggested by the doctor.

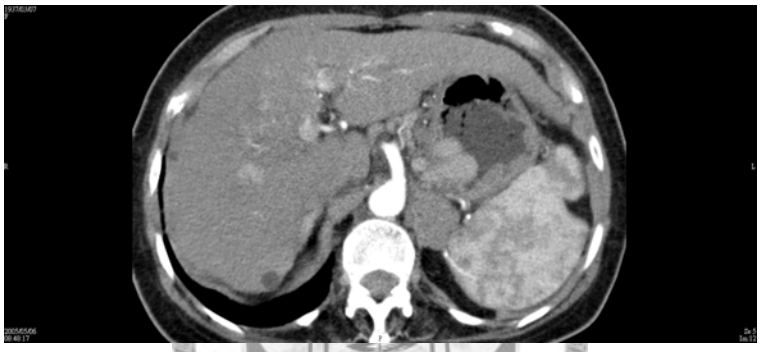


Figure 17 Pre-NK multiple hepatic tumors with portal vein thrombosis

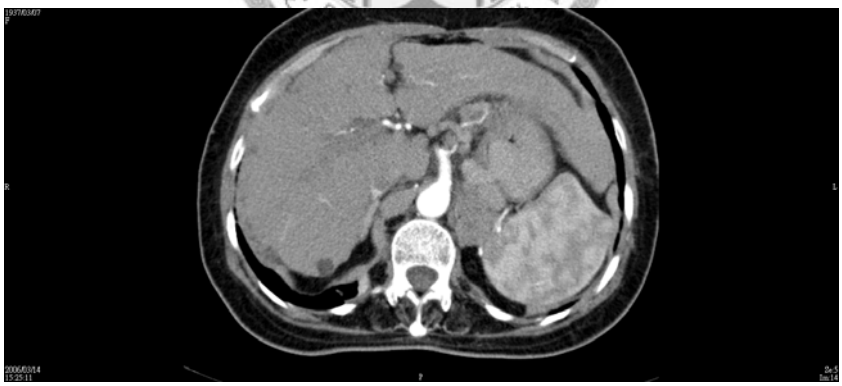


Figure18 Post NK cancer totally disappeared

Method:

She entered the clinical trial of autologous NK cell therapy against malignancy.NK cells was ex-vivo cloned under GLP supervision.

Immune cell profle to be checked by backscattered signal of two-clor

flowcytometry.

Course:

She received more than 70 times autologous NK cell transfusion in 4 years.

She

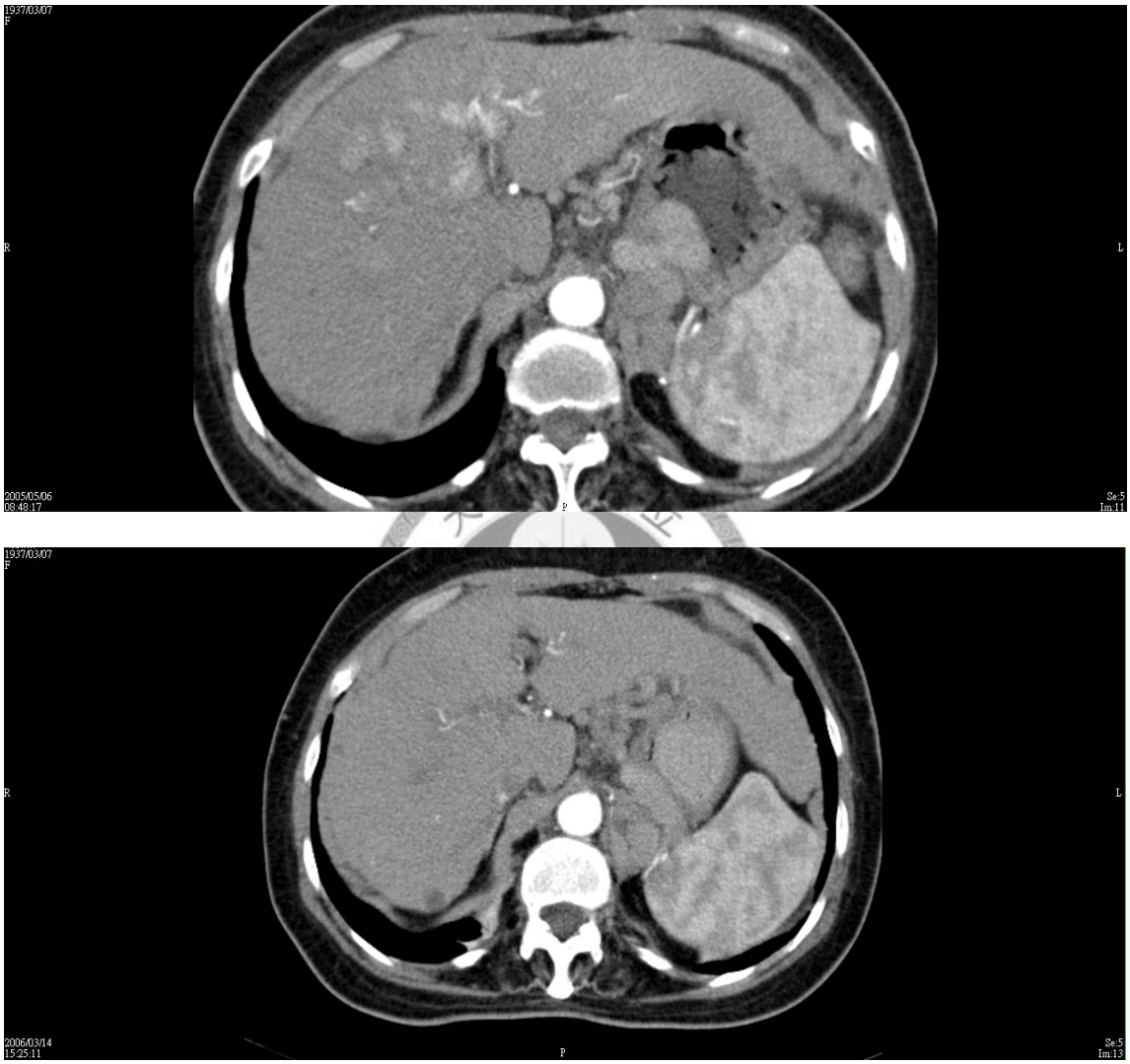


Figure 19.20Similar pre and post findings

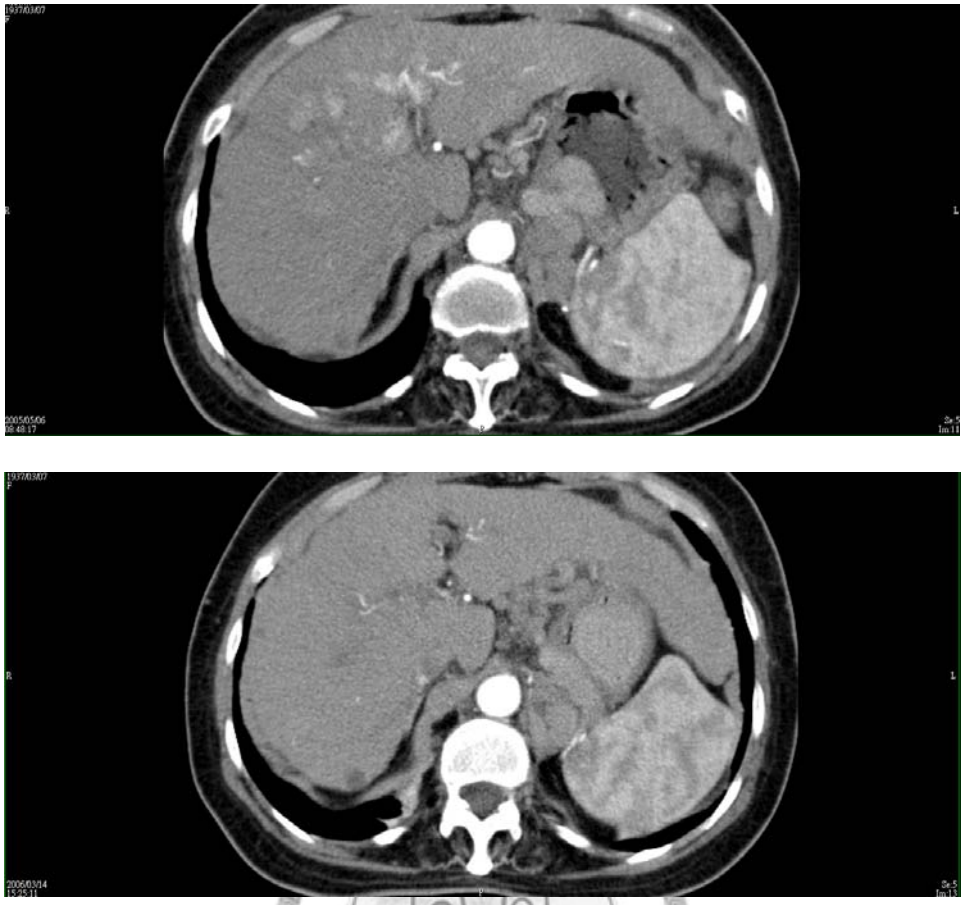


Figure 21.22Similar pre and post finding

received no other treatment. During the course, she led a un-eventful life, and still kept her daily resource recycling volunteer job in Tsu-Chi.

Results:

Good appetite,good activity,no pain all the time in the course. Her BMI was the same all the time. The HCC and liver cirrhosis show no disease progression, Her liver enzyme GPT from above 100 back to norma valuel. And miraculously,.Her HCC and portal thrombosis disappeared. She survives till now for four years.

Discussion:

In this case study, NK cell therapy is the only therapy. In three aspect NK could have its effect: 1.NK kills the cancer cells, evidenced by the disappearance of

tumors and portal vein thrombosis, it is the innate immunity per se.against cancer.
2.NK cell easy the liver inflammation, evidenced by the return of liver enzyme GPT.
3.NK fight against the viral infection.

This is a proof to the proposed immune-inflammatory model of human longevity
and risks in several ways. Eliminate the most severe disease, terminal malignancy, could be achieved by our innate power, the immunity. Ease of the inflammation is a prodromic sign of the return of human health. Evidenced by so uneventful the course, no cachexia , active, no pain, no weakness, no need to live in a hospital, no medication, in other word ‘ **healthy**”.

And ease of inflammation is a possible way to relieve the longevity risks. In malignancy, it is well documented that inflammatory cell colonized in cancer tissue. Inflammation leads to cancer by the accumulation of Treg, macrophage, dendritic cell which help to escape from the immunesurveilance.

The success of this case should be interpretated carefully. In the following case studies. NK cell s do not necessarily kill all the cancer tissues. Author will recommend combination therapy with other modality like radiation therapy or HIFU(high intensity focused ultrasound) after detail search through biosignals generated by inflammation and cancer.

5.4 NK Case Study 2 Lung Cancer Stage 4

Background

64 year old gentleman, ex-smoker for 40 years, visited my clinic with diagnosis of Lung adenocarcinoma, stage4.

There was originally a 3 cm x 3 cm right lung lower lobe tumor, with

metastasis to neck LN x 1, mediastinum LN x 2, brain metastasis x2, scapular metastasis x 1 Neck LN was removed. Chemtherapy had been done by paclitaxel for 2 courses. Radiation therapy 30 times had been done on the lesions in, mediastinum, brain and scapula. Residual tumor 1cmX1cm in lung and scapula lesion remained . Patient the was on Tarceva 1# qd.for 3 month untolerable, so he discontinued 1 month after NK started. He received NK transfusion for 32 times.in 3 and a half years.

Method and Course

Scapula lesion disappeared.. He has no cough, no dyspnea, no pain, activity was good. His body weight remains the same. He remained progression free for 3 years. No constitutive symptom of cancer. He lengthened the interval of NK transfusion to once every 6 weeks. He did not achieve complete remission. Lung lesion remains. He moves to Veteran General Hospital pursuing eradication.

Results;

NK cells toghther with other therapy make this case progression free for 3 years with no need for hospitalization.

Discussion:

The innate immune NK cell still have effect on survival and progression-free interval. Compared to similar Tarceva treatment the average survival is 6.7 months: progression- free survival 2.23 months. Less than 25 percent suvives 2 years. This case has progression-free survival for 3 years and is still fighting against his cancer.

Conclusion:

NK cell therapy in this lung cancer study is effective in prolonged progression-interval, prolong survival. The patient survives 46 months and still leave an uneventful life despite his cancer is still reach complete remission.

5.5, NK Case Study 3 Pancreatic Cancer Stage 4

This 70 year-old Chinese gentleman was found to have pancreatic head cancer 4.5cm x 2.8 cm during an abdominal ultrasound examination. After clinical staging, stage 4 pancreatic cancer with numerous liver metastasis and direct aorta invasion was diagnosed. The tumor was unresectable due to direct great vessel invasion. The attending physician felt that this gentleman would not have a quality survival with traditional surgical and chemo- therapeutic and persuaded the gentleman into radiation therapy to pancreatic head. The radiation therapy was more palliative than curative.

Radiation therapy against the pancreatic head tumor was performed for 24 times. At mean time, NK transfusion has been started for more than 40 times. 3 months afer start of both treatments, tumor of pancreatic head shrank from 4.5cmx2.8cm to 2.8cmX 2.0cm, and the extent of aorta invasion diminished. Follow up check up by CT show tumors stable in number and size and invasiveness. 24 month after diagnosis of stage 4 disease patient is totally no symptom, no jaundice, no need for hospitalization.

Early pancreatic cancer was usually symptom-free. With tumor growing, upper abdominal pain, back pain, jaundice, weight loss, diarrhea will occur in less than 6 months. And the survival is extremel poor. Patients who are initially diagnosed with metastatic pancreatic cancer (stage4) have a poor prognosis, with survival averaging only three to six months despite treatment. This patient survives well , and still lead a life with good quality. He even traveled to Japan, Russia with no paticular attending. His terminal pancreatic cancer is progression-free for 24 months and completely symptom free. He has very good appetite, keep body weight well and even gain three kilograms, and develops no emaciation. The course is so unusual in progression free, symptom-free, emaciation- free that the efficacy of the NK transfusion based

combination therapy could not be ruled out.

5.6 NK Case Study 4 Adrenal Gland Cancer Stage 4

Eight years ago, this 53 year-old Singapore gentleman was diagnosed as adrenal gland cancer stage 4 in the United States of America. The Adrenal gland cancer, left, was 10cmX 10cm with direct left liver lobe invasion and numerous liver metastasis to bot liver lobes, and one brain metastasis.

The Adrenal gland tumor and a part of left lobe of liver was surgically removed. Chemotherapy was given, which was very tortuous and with a lot of complication. He spent intermittently for six months in one and a half years and struggled to come back to Asia.

He came to our China affiliation and received NK transfusion about 6 year ago . Combination therapy with a full course radiation therapy against the brain metastasis was performed with complete remission. And the liver tumors showed progression-free and some tumor decreased in size. Transarterial embolization was done to those liver tumors with accessible artery without complication. The management of three residual tumors remains pending for he has only about half size liver remains.

He receives about a hundred times NK tansfusion in 6 years. Despite his residual malignant tumor, he has no symptom, no emaciation, no complications, farly contrast to chemotherapy with complications and side effects. He had several hospitalizations for treatment not for complications. The cumulative hospitalization days after NK transfusion is less than 20 days. Otherwise, his life is uneventful. He survives stage 4 malignancy for eight years is still alive well. He pursues eradication of the residual tumor after more normal liver tissue regeneration.

Chapter 6 Conclusion

In the beginning of the thesis, we ask ourselves three questions:

Is it possible to treat diseases, like cancer, through the inner power of our own?

What is the extent that immunity can help ?

Is it possible to prevent diseases and achieve longevity through the power of our own?

To which the answers are pretty positive.

We propose our theory idea of unified model of longevity and diseases, named as an immune –inflammatory model of human longevity and risks. Pro-inflammatory cytokines, Inflammation, and inflammatory cells are tightly tied with disease pathology in cancer atherosclerosis, ageing, organ degeneration, and ageing- related diseases. Make inflammation as very important common pathway and major risk leading to diseases, and most morbidity and mortality. Further make anti-inflammatory measure good candidate to slow or prevent diseases.

The case study on cancer innate immune therapy proves that it is possible and reasonable to use our own innate immunity to treat cancer. Autologous natural killer(NK) cell transfusions eradicate cancer in pilot study. Terminal hepatocellular carcinoma with portal vein thrombosis totally disappeared with NK cell transfusion as sole treatment. NK cell transfusion prolongs survival in all cases. NK cell transfusion lengthens symptom-free intervals and progression free intervals. It is freely used in any combination therapy. The life quality of patient is much superior to otherwise treated patients. Here We answer the question:

Is it possible to treat diseases, like cancer, through the inner power of our own?

"Yes, we may treat cancer, the most notorious diseases, through our own innate immunity.' We prove it by NK case study to demonstrate effect in survival, and mobidity, motality lowering. There are immediate two questions

What is the extent that immunity can help ?

Is it possible to prevent diseases and achieve longevity through the power of our own?

In the following, we are trying to answer them.

The phenotype change toward NK cell dominance is not only useful in treating cancer is is also useful in prevention against atherosclerosis and ageing-related conditions.The phenotype of NK cell(natural killer cell) CD 56 dim dominance people was Coronary Heart Diseases exempt. On the contrary, coronary heart disease patients has phenotype of less NK cell. NK is the immune phenotype of healthy longevity. Those with higher NK has favorable endocrine profile, more muscle , more life activity, and less morbidity.

Anti-inflammatory measures are also good candidates to slow the onset of cancer, atherosclerosis, and many ageing –related diseases. Non-chemical approach are emphasized in diseases prevention model: unload the immune by elimination of food antigen, elimination of food toxins ; and unload the mental stress. For its non-chemical non-invasive characteristic is a feasible approach for every one in daily life to decrease the health risks.

In inflammation case study, we test electrical bio-feedback as a non-invasive non-chemica measure to fight against inflammation. The nature of non-invasiveness and the efficiency of this approach strengthens the basis for

using electrical bio-feedback in disease prevention in non-hospital setting.

NK treats and prevents cancer. NK prevents atherosclerosis. NK cell treats variety of infection and brings inflammation back to homeostasis. NK is the phenotype of healthy ageing. There is easily available abundant NK precursor cells in the peripheral blood . Make NK proliferation technology very important subject in this immune- Inflammatory model of human longevity and risks.

. Thus, we conclude that having your life immune-friendly way, lowering the inflammation to prevent diseases, selecting effective and immune-harmless treatments against diseases, and promoting or changing into NK prevalent immune phenotype, would be the key for the human being to live beyond the current limitation of longevity.

REFERENCE

1. Bhardwaj N, Harnessing the immune system to treat cancer J. Clin. Invest., May 2007; 117: 1130 - 1136

2. Chidrawar S M , Khan N, Tracey Chan Y L, Nayak L and Moss P.. Ageing is associated with a decline in peripheral blood CD56bright NK cells Immunity & Ageing,2006: 3:10 doi:10.1186/1742-4933-3-10 2

3. Franceschi C, Bonafe M, Valensin S, Olivieri F, De Luca M, Ottaviani E, De Benedictis G: Inflamm-aging. An evolutionary perspective on immunosenescence. Ann N Y Acad Sci 2000, 908:208-218.3.

4. Franceschi C, Bonafe M: Centenarians as a model for healthy aging. Biochem Soc Trans 2003, 31:457-4614.

5. Hak L , Myśliwska J , Więckiewicz J, Szyndler K, Trzonkowski P., Siebert J. and Myśliwski A..NK cell compartment in patients with coronary heart disease. Immunity & Ageing , 2007: 4:3, doi:10.1186/1742-4933-4-3

6. Kärre, K., Ljunggren, H. G., Piontek, G. & Kiessling, R. Selective rejection of H-2-deficient lymphoma variants suggests alternative immune defense strategy. Nature 1986: 319: 675–678 .

7. Levy, S.M., Herberman, R.B., Lee, J., Whiteside, T., Beadle, M., Heiden, L., Simons, A., Persistently low natural killer cell activity, age, and environmental stress as predictors of infectious morbidity. Nat. Immun. Cell Growth Regul. 1991:10: 289–307.

8. Licastro F., Candore G., Lio D., Porcellini E., Colonna-Romano G., Franceschi C. and Caruso C., Innate immunity and inflammation in ageing: a key for understanding age-related diseases, Immun. Ageing 2005 : 2: p. 8.

9. Lombardi F. Sympathetic activation and sub-clinical inflammation: a new

combination to identify high risk subjects. European Heart Journal 2004: 25: 359–360

10. Mariani, E., Ravaglia, G., Forti, P., Meneghetti, A., Tarozzi, A., Maioli, F., Boschi, F., Pratelli, L., Pizzoferrato, A., Piras, F., Facchini, A., . Vitamin D, thyroid hormones and muscle mass influence natural killer (NK) innate immunity in healthy nonagenarians and centenarians. Clin. Exp. Immunol. 1999116, 19–27.
11. Pawelec G, Solana R, Remarque E, Mariani E: Impact of aging on innate immunity. J Leukoc Biol 1998, 64:703-712.
12. Sajadieh A, Wendelboe Nilsen O, Rasmussen V, Hein HO, Abedini S, Jorgen FH. I increased heart rate and reduced heart rate variability are associated with subclinical inflammation in middle-aged and elderly subjects with no apparent heart disease. Eur Heart J 2004;25:363–70.
13. Troen B.R., The biology of aging, Mt. Sinai. J. Med. 2003: 70 :pp. 3–22.
14. Plackett TP, Boehmer ED, Faunce DE, Kovacs EJ: Aging and innate immune cells. J Lekoc Biol 2004, 76:1-9.
15. Shiu-Nan Chen, Application of Mushroom Immunomodulatory Peptides, β-Glucans and Fibrinolytic Enzymes on Cancer and Cardiovascular Diseases, 2008
16. Hanna N. The role of natural killer cells in the control of tumor growth and metastasis. Biochim. Biophys. Acta, *780:* 213-226, 1985.
17. Wiltrout R. H., Herberman R. B., Zhang S. R., Chirigos M. A., Ortaldo J. R., Green K. M. J., Talmadge J. E. Role of organ-associated NK cells in decreased formation of experimental metastases in lung and liver. J. Immunol., *134:* 4267-4275,
18. Shivani M Chidrawar, Naeem Khan, Y L Tracey Chan, Laxman Nayak, and Paul AH Moss Ageing is associated with a decline in peripheral blood CD 56

bright NK cells Immun. Ageing 2006 : 3: p.108. Published online 2006 November 29. doi: 10.1186/1742-4933-3-10

19. Jen-Chien Chien, Jeng-Pang Wang, Chien-Lung Cho, Fok-Ching Chong, "Security Biosignal Transmission based on Face Recognition for Telemedicine," *Biomedical Engineering-Application,Basis & Communications*, 19 (1), 63–69, 2007 (SCI)
20. Jeng-Pang Wang, Yaw-Jen Lin, Fok-Ching Chong, " A Study on Component-Centric Document Database Architecture in HL7 and CDA Applications "Joint Conference: 2007 IEEE Healthcom/HL7 AP/IPv6 Summit, Taipei 2007
21. Jeng-Pang Wang, Fok-Ching Chong, "An Computational Area Approach to Evaluate Flow-mediated Dilatation," 27th the World Congress on Medical Physics and Biomedical Engineering 2006(WC2006), Seoul, Korea, 2006
22. Chien Jen-Chien, Wang Jeng-Pang, Hsueh Meng-Lun, Cheng Po-Shuen, Fok-Ching Chong, ""An Effective Image Measurement in Brachial Flow-Mediated Dilation Response Analysis," 27th Annual Int. Conf. IEEE Eng. in Med. & Bio. Soc., Shanghai, China, Sept. 2005
23. Jeng-Pang Wang, Fok-Ching Chong "A Real-Time Wireless Physiological Monitoring System for Nursing Center" Progress In Electromagnetics Research Symposium, Hangzhou, August. 2005
24. Min-Yee Chen, Fok-Ching Chong"A Study of Heart Rate Variability and Blood Oximeter and Sleep Apnea" : matser theisis of graduate institute of electrical engineering, National Taiwan University (2007).

PUBLICATION LIST

1. Jeng –Pang Wang, Jen-Chien Chien, Chin-Dr Fan, Cheng-Chin Huang, Cheng-Yu Fan , Fok-Ching Chong , Yaw-Jen Lin “Construct an Approximation Decision Model of Medical Record by Neural Networks for Clinical Diagnosis Guideline-The Ophthalmology Department as an Example” Biomedical Engineering-Application,Basis & Communications, 2009 (Accepted) (SCI)
2. Jen-Chien Chien, Jeng-Pang Wang, Chien-Lung Cho, Fok-Ching Chong, “Security Biosignal Transmission based on Face Recognition for Telemedicine,” Biomedical Engineering-Application,Basis & Communications, 19 (1), 63–69, 2007 (SCI)
3. Yaw-Jen Lin, James Wang, Chia-Feng Su, Heng Shen Chen, Fok-Ching Chang, “A Study on Constructing SCORM-compliant e-Learning Teaching Materials from CDA Documents,” The Journal on Information Technology in Healthcare, 3(2), 101-108, 2005 (EI)
4. Yaw-Jen Lin, James Wang, Nick Shu, Fok-Ching Chang, “Construct Teaching Materials from HL7/CDA Electronic Patient Records according to SCORM Standard forMedical Education” Biomedical Engineering-Application,Basis & Communications, 16(4) 190-198, 2004(SCI)
5. Jeng-Pang Wang, Yaw-Jen Lin, Fok-Ching Chong, “ A Study on Component-Centric Document Database Architecture in HL7 and CDA Applications “Joint Conference: 2007 IEEE Healthcom/HL7 AP/IPv6 Summit, Taipei 2007
6. Jeng-Pang Wang, Fok-Ching Chong, “An Computational Area Approach to Evaluate Flow-mediated Dilatation,” 27th the World Congress on Medical Physics and Biomedical Engineering 2006(WC2006), Seoul, Korea, 2006
7. Chien Jen-Chien, Wang Jeng-Pang, Hsueh Meng-Lun, Cheng Po-Shuen, Fok-Ching Chong, ““An Effective Image Measurement in Brachial Flow-Mediated Dilation

Response Analysis," 27th Annual Int. Conf. IEEE Eng. in Med. & Bio. Soc., Shanghai, China, Sept. 2005

8. Jeng-Pang Wang, Fok-Ching Chong "A Real-Time Wireless Physiological Monitoring System for Nursing Center" Progress In Electromagnetics Research Symposium, Hangzhou, August. 2005
9. Jeng-Pang Wang , Dershing Luo , Meng-Lun Hsueh, Fok-Ching Chong and Jan-Pine Sun, "A Survey in Deploying e-Learning Systems in Taiwan's Medical Education," 1st International Conference on Capacity Building for Information Technology Integration in Teaching and Learning, Hong Kong June, 2005(CBIT2005)