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Development of Blood Tests For Early Cancer Detection

Brian Schaefer, D.Phil. (Oxon)

Going Back To Our Roots

This months featured herb is the dandelion (Taraxacum Officinale)

Phytotherapy News

Reishi Mushroon Active Identified

THIS ISSUE

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PHYTOTHERAPY NEWS

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Reishi mushrooms have been used in traditional chinese medicine for thousands of years

COVER STORY

Development of Blood Tests for Early Cancer Detection

The development of these blood tests for early cancer detection and monitoring build upon two central bodies of research – research into the anti-cancer activities of the phytonutrients known as Salvestrols and the research into the metabolic activity of the enzyme CYP1B1

UPFRONT

To study the prevalence of sedentary behaviours and decreased physical activity among school going adolescents in Ludhiana

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EDITORIAL

Professor Gerry Potter
Editor

Development of Blood tests for Early Cancer Detection

The main feature article of this edition of IJOP describes research aimed at developing a test for cancer that will allow much earlier diagnosis than is presently available. The article describes two new cancer diagnostic blood tests. The first directly measures the levels of the CYP1B1 protein in the bloodstream. In normal volunteers the level of CYP1B1 is vanishingly small but this level is highly elevated in cancer patients which makes it useful for early cancer diagnosis. The second test measures the salvestrol metabolites that result from CYP1B1 activity. The levels of unreacted salvestrol and activated salvestrol metabolites are measured in the bloodstream and this gives an indication of how well the salvestrols are working by providing evidence for their metabolism.

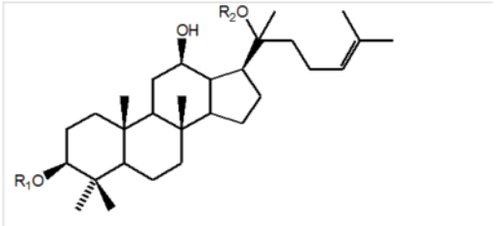
This months IJOP includes a guest article from the College of Physiotherapy in the city of Ludhiana in Northern India. This article presents evidence for a concerning trend in increased sedentary behaviour of adolescents in India, and shows that the developing world is following the western world in this trend which can lead to an array of health problems later in life.

Professor Gerry Potter

Ginseng Research

Phytotherapy Research News

Panax Ginseng is one of the most extensively studied medicinal plants, and the roots of ginseng contain a multitude of bioactive molecules each with their own pharmacological action. The main bioactive compounds are triterpene glycosides known as ginsenosides. There is a mysterious duality to the actions of ginseng which are likened to a ying yang of opposites. On the one hand ginseng can help with wound healing which involves angiogenesis, and on the other hand ginseng has anticancer effects due to its anti-angiogenic action. How can one plant have these two opposite actions. Well it turns out there are two different molecules responsible for these actions one of which is pro angiogenic and the other is anti-angiogenic. These are both ginsenosides of the panaxadiol family and differ mainly in the positions of the glucose goups. The pro angiogenic compound is ginsenoside Rg1 which is a panaxatriol glycoside, and works by binding to a human glucocoticoid receptor to elicit an angiogenic response and aid in wound healing.

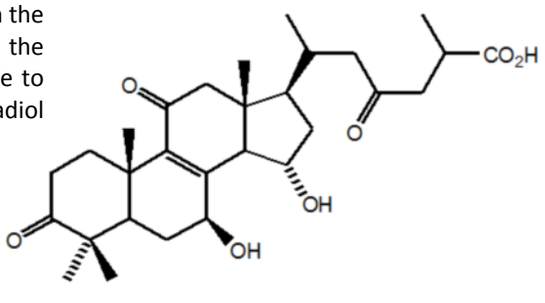


Ginsenoside Rb1; R1 = Glu-Glu, R2 = Glu-Glu
Compound K; R1 = H, R2 = Glu

The anti-angiogenic compound has been identified as ginsenoside Rb1 which is a panaxadiol glycoside, and this works by binding to the human estrogen receptor ER-beta (but not ER-alpha) where it acts as an agonist to promote ER-beta DNA transcription and protein expression. This leads to the ER-beta stimulated production of the powerful natural anti-angiogenic protein PEDF. This is the most potent anti-angiogenic protein expressed by the human body and so the Rb1 stimulated production of PEDF accounts for its antiangiogenic activity. This reasearch shows the importance of the glucose carbohydrate groups in eliciting biological function. However the ginsenoside glucosides are modified by intestinal bacterial action which involves the sugar moieties being removed. The ginsenoside Rb1 is a tetra-glucose derivative that is cleaved to the mono-glucose derivative compound K that is the active metabolite that passes through the intestinal wall and enters the bloodstream. What is interesting here is that the ginsenoside mono-glucose conjugate is able to be absorbed with the sugar molecule intact into the bloodstream and this is probably due to the high lipophilicity of the panaxadiol triterpene group.

Reishi Mushroon Active Identified

Reishi mushrooms have been used in traditional chinese medicine for thousands of years and are among the more popular alternative treatments for cancer. Even though the anticancer activity of Reishi mushrooms is acknowledged by alternative therapy practitioners it has taken until now for scientists to identify the active component. Recent research has lead to the identification of the active compound Ganoderic Acid, named after the Reishi mushroom genus *Ganoderma Lucidum*. Ganoderic Acid is an oxidised derivative of the triterpene lanosterol and is the fungal metabolite that has resulted from lanosterol oxidation. This compound has carbonyl goups at the 3 and 10 positions of the steroid which mimic cortisone. Ganoderic Acid has been found to inhibit the enzyme Topoisomerase which is the target of the clinically used anticancer agents Topotecan and Irinotecan.



Ganoderic Acid from Reishi Mushrooms

FEATURED HERB

Featured Herb – Dandelion

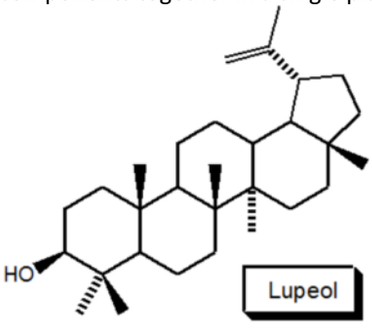
Going Back To Our Roots

This months featured herb is the dandelion (*Taraxacum Officinale*) which is part of the Compositae family of plants. This family has two sub-families known by their trivial names as the “Daisy Family” which includes many yellow flowered plants such as sunflower, yarrow and tansy, and the “Thistle Family” which are purple flowered and includes milk thistle, burdock and artichoke.

The diuretic properties of dandelion leaves are well known historically but it took until 1978 for this effect to be clinically proven. Dandelion has been used traditionaly to treat various diseases including cancer, and one of its traditional names is “Cankerwort”. Phase 1 clinical trials of dandelion root tea for treating cancer have just started in Canada following observations of its beneficial activity. This is an important development and is one of only a few examples of a natural product entering clinical trials.

There are 3 classes of molecules in dandelion that have anticancer properties, the sesquiterpene lactones, the triterpenes, and the salvestrols. The sesquiterpene lactones such as taraxinic acid are toxic compounds and are the plants “don't eat me” molecules. These are the anti feedants that deter grazing animals from eating them. The lactone ring contains an acrylate group which is an alkylating agent that can react with proteins and DNA bases causing toxicity. In this respect they are similar to the anticancer alkylating agents used in chemotherapy such as Melphalan. These molecules are mopped up by glutathione in the liver and these molecules elicit a detoxification response which involves expression of glutathione by the liver and increased urination resulting in its diuretic effects. These molecules are mainly present in the leaves and gives them their characteristic bitter taste. The young leaves have lower levels and can be safely eaten and added to green salads.

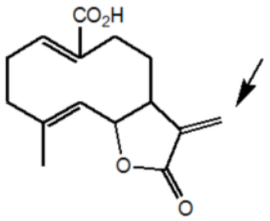
As a food dandelion leaves and roots are a source of vitamins, minerals and carbohydrate. The vitamins present are pro-vitamin A (carotene), vitamin B3 (niacin) and vitamin C. The minerals present include potassium, calcium and magnesium. The carbohydrates present are short polymers of glucose and fructose in the form of Inulin. It is interesting that dandelions contain both the salvestrols and their co-factors such as niacin and magnesium which are required for their biological activation, so mother nature has formulated all these components together in a single plant.



Lupeol

The triterpenes found in dandelion include lupeol, taraxasterol, *beta*-sitosterol, and stigmasterol. These are made by the plants using a complex synthetic route involving squalene cyclisation, by a process similar to that used by the human body to make the human triterpenes lanosterol and cholesterol. These plant derived triterpenes can interfere with cholesterol biosynthesis and have the ability to lower cholesterol levels. Lupeol has recently caught the attention of researchers since it is able to induce apoptosis in tumour cells without causing normal cell toxicity in a similar manner to the salvestrols. Lupeol is also found in exotic fruits such as mangoes. Lupeol is related to betulin found in the silver birch that also has pro-apoptotic activity. The triterpenes and salvestrols are found mainly in the dandelion roots and are present as their sugar glycoside derivatives.

Taraxinic Acid



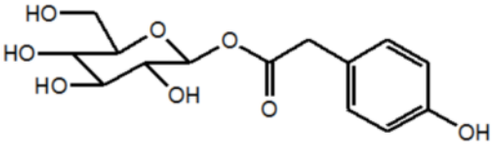
A fourth class of molecules is present in dandelions that have anti-inflammatory activity which are the phenolic acid gylcosides such as Taraxacoside. This is a glucose conjugate of 4-hydroxy phenylacetic acid which is similar to the non steroidal anti-inflammatory drugs Diclofenac and Ibuprofen.

Dandelion roots are a rich source of salvestrols and these are now thought to be the principal contributors to its anticancer activity. The dandelion features in traditional herbal medicine manuscripts and is found in the Welsh Herbal written in the 12th century. Culpepper describes dandelion as good for increasing urinary flow, and for healing inner ulcers. Chevallier's “Encylcopedia of Medicinal Plants” describes dandelion root as having powerful anti-inflammatory properties useful for the treatment of eczema, psoriasis, and arthritis, and these are all conditions which can respond to treatment with salvestrols. In traditional chinese medicine (TCM) dandelion root is known as Pu Gong Ying and has been used in China for breast cancer therapy.

I like the dandelion and admire its resilience and ability to survive in the wild. It has a cheerful yellow flower that lifts the spirits and brightens up our lawns. They do not need watering or fertilizers and happily grow without any human assistance. They are a gift from mother nature which are freely available and are an important component of the herbal medicine cabinet.

Recipe for Dandelion & Burdock Tea

Take 5 tsp of dandelion root and 2 tsp of burdock root and place in a saucepan. Add 250 ml of boiling water and simmer for 20 mins. Allow to cool then strain through a sieve into a jug ready to drink.



Taraxacoside

UPFRONT

To study the prevalence of sedentary behaviours and decreased physical activity among school going adolescents in Ludhiana

By Sanjeev Kumar Khanna.[1] Avnee Sarin [2] K.E.Benjamin .[3]

ABSTRACT

Objective: To study the prevalence of sedentary behaviours and decreased physical activity among school going adolescents in Ludhiana.

Material and Method: School going children of Ludhiana of age group 13-17 years were enrolled in the present study. All subjects were given a Questionnaire for this survey which was filled and completed by them and their leisure time spent in sedentary and non-sedentary behaviour per day during weekday and weekends was collected and then collected data was analysed.

Result: 100 school going students of Ludhiana, between age group of 13-17 years were taken under the inclusion criteria. The result of the study showed that the students spent 266 minutes (4.43 hours) on average in sedentary activity and only 15 minutes were spent in non – sedentary behaviours on weekdays and on weekends time spent on sedentary activity was increased to 275 minutes (4.58 hours)in sedentary activity and 15 minutes in non- sedentary one.

Conclusion: This survey has concluded that there is prevalence of sedentary lifestyle and decreased physical activity among school going adolescents. Our sample showed that there was very low level of leisure time physical activity among school going adolescents as recommended by W.H.O. So, there is major need to promote leisure time physical activity among adolescents to have a better health in future, prevent obesity and other health related problems.

Keywords: sedentary lifestyle, non- sedentary lifestyle, Ludhiana school going adolescents, leisure time sedentary activity questionnaire.

BACKGROUND

A sedentary lifestyle is the medical term used to denote a type of lifestyle with no or irregular physical activity.[1][2] Sedentary activities include sitting, reading, playing certain videogames, using the computer, watching television, etc. for much of the day with little or no vigorous physical exercise.[2]

Physical activity is a fundamental means of improving physical and mental health.[3]

Adolescence is the period of psychological and social transition between childhood and adulthood. The health habits and coping skills developed during this period carry into adulthood. Adopting adequate physical activity levels during childhood and adolescence is important to prevent chronic disease later in life. [4] Regular participation in physical activity is an essential component of a healthy lifestyle. [4] Among adolescents regular physical activity actually reduces the onset of obesity, the number of cardiovascular diseases and is positively associated with physical fitness.[4] Furthermore, there are strong evidence that sedentary behaviours adopted during childhood track into adult life.[5]

Recommended guidelines for physical activity and time spent in other sedentary activities say that children and youth should accumulate at least 60 minutes of physical activity on a daily basis.[6] Extended period of time (2 hours or more per day) spent on sedentary pursuits (TV viewing, computer use , etc.) are associated with decreased physical activity levels and increased obesity and overweight.[6]

The present study, helps us to know individual students leisure time sedentary and physically active behaviours.

Materials and Methodology-

It was a Survey study conducted on a school students with total sample size of 100, between age group of 13-17 years. Stratified Random Sampling technique was used.

Hypothesis

Null Hypothesis: The Children have adequate amount of Physical Activity and less sedentary behaviour.

Alternate Hypothesis: The Children have decreased level of Physical Activity and more sedentary behaviour.

Variables:

Duration of participants physical activities (sports, active transport and other habitual physical activity) and sedentary activities (TV viewing, computer use sit and talk etc) in leisure time was converted into minutes.

Instruments:

Leisure time physical activity questionnaire for adolescents .

Procedure-

Firstly, questionnaire was prepared which included close ended questions only. Then a pilot study was done on 20 students to

check the validity and reliability of the questionnaire by intertester and intratester methods. Questions were easily understood and well answered by subjects.

After the successful completion of pilot study the questionnaire was distributed among 100 students. Each subject is given verbal introduction for the questionnaire. A signed consent form was obtained from each subject prior to participation in the study. After this subjects were made to fill the explained questionnaire and are given instructions to tick the option which most closely relates them. After getting back the Questionnaires data was analysed and results were obtained.

DATA ANALYSIS: Data was analysed in Microsoft excel 2007 by calculating average, percentage, mean, median and mode method and final results are formed which are as under pie graph 1 and 2. Table 1 shows the average mean time spent in leisure sedentary and non-sedentary behaviour during weekdays and weekends.

FIG: 1 a (on the next page) Shows average time spent on sedentary activity was 266 minutes (4.43 hours) per day with +/-63.3 SD and 275 minutes (4.58 hours) per day +/- 69.8 SD weekdays and weekends respectively.

Time spent on non-sedentary activity was 15 minutes per day +/- 10.2 SD on weekdays and 15 minutes +/- 15 SD on weekends.

TABLE: 2a shows per question the average mean time spent

Fig: 2 a Shows students leisure time spent on different activities both on weekdays and weekends. The graph shows doing homework, taking tuitions and reading books occupied most of the leisure time i.e. average 90 minutes(1hour 30 minutes) per day both on weekdays and weekends. After this T.V. viewing, listening to music, sitting and talking with family and friends was the most common activity.

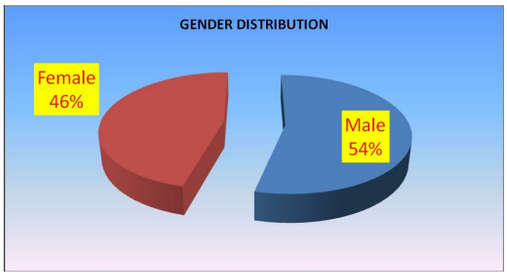
The other commonly done activity was using computer and playing videogames and other sedentary activities of arts. The average time shows that using computer, playing videogames and T.V. viewing activity was more on weekends than that of weekdays.

The graph also showed that usage of motorized transport was more on weekdays than weekends i.e. on weekdays average 42 minutes spent per day.

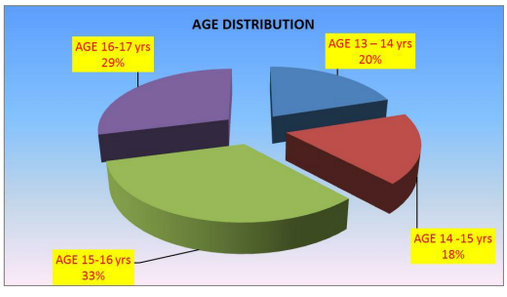
The least time was spent on playing sports, outdoor games, exercises, other physical hobbies and active transport.

Fig: 2 b Shows total screen time was 133 minutes (~2hours) per day and 161 minutes (~ more than 3hours) per day on weekdays and weekends respectively.

PIE GRAPH 1



PIE GRAPH: 2



DISCUSSION

This study reports descriptive data on physical activity and sedentary behaviours of adolescents. Using questionnaire method enabled us to get information on time spent on various activities during leisure time.

In our sample as in other data sets from Australia[7], USA[8], UK[9-10], Hungary[11] and elsewhere[12], TV viewing was the most popular sedentary behaviour in leisure time. Estimates from the present sample are similar to those from other studies[12], with more than 1hours of TV viewing for weekdays and above 2 hours on weekends. There is broad agreement that 4 hours of TV viewing per day is excessive[13] and that < 2 hours per day are recommended. With the development of new technologies screen time should be focused (TV, computer, videogames etc.). In our study in addition to TV viewing students spent ~1hour on weekdays using computer and playing videogames and >1hour on weekends. This appears to be similar estimates for other countries when averaging across a 7 day week.[12]

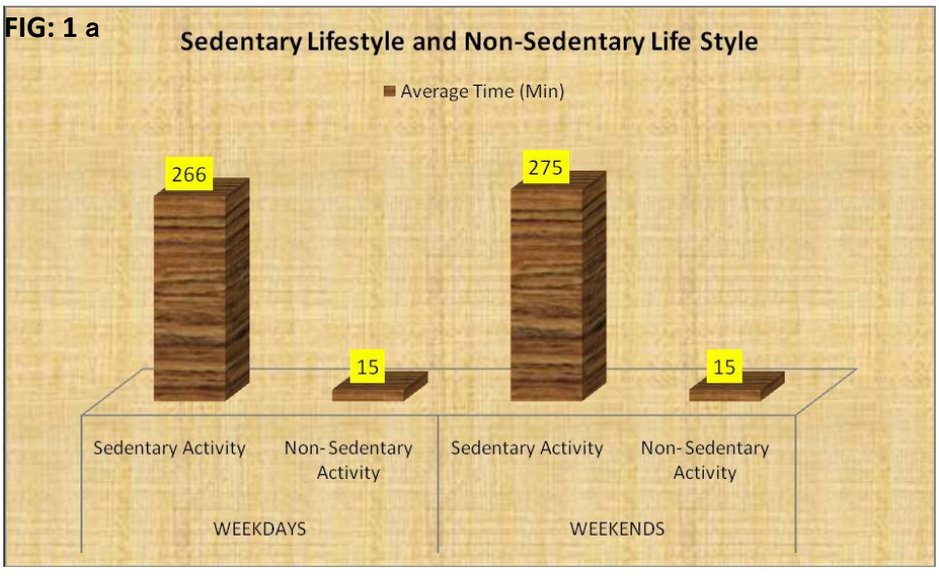
Table 1

TIME	WEEKDAYS		WEEKENDS	
	Sedentary Activity	Non- Sedentary Activity	Sedentary Activity	Non- Sedentary Activity
Average Time (Min)	266	15	275	15
SD	63.3	10.2	69.8	15

TIME [Average Time in Min]	WEEKDAYS		WEEKENDS	
	Sedentary Activity	Non- Sedentary Activity	Sedentary Activity	Non- Sedentary Activity
Male	259	16	281	13
Female	274	14	269	18

TIME [Average Time in Min]	WEEKDAYS		WEEKENDS	
	Sedentary Activity	Non- Sedentary Activity	Sedentary Activity	Non- Sedentary Activity
13 – 14 yrs	259	15	289	17
14 -15 yrs	259	13	268	16
15-16 yrs	268	15	285	15
16-17 yrs	273	16	260	14

FIG: 1 a



This total screen time for weekdays in the sample was approximately 2.2 hours. and for weekends 3 hours or more per day. So it is important to help young people monitor the amount of their time spent on screen behaviours as some will be having excessive levels. The data suggests that prolonged sitting time, typical of screen use may be associated with unfavourable metabolic profiles.[14] So, breaks in the sedentary time[15] and more time in moderate or moderate to vigorous physical activity are recommended.

At the same time our data showed that non screen based sedentary behaviours are also prevalent. These include motorized transport, doing homework (most common) etc. This suggests higher levels than in some other countries.[16]

Scully and colleagues,[17] for example highlighted that Australian state education department guidelines suggest , that students complete about 10 minutes of homework a day each school year, they progress in school to a minimum of 2 hours per day in year 12. Homework therefore is a significant sedentary pursuit. Being sedentary for transport is one area where policy and promotion can be effective. There is clear competition between active and motorized modes of transport. For students living close to their school there is good opportunity to take more active forms of transport and to offset the decline in walking and cycling that is seen in many countries.[18] For example, in our sample 80% of the students take no active transport at weekends, suggesting that a great deal of their time may be spent at home. Time inside home is predictive of more sedentary behaviour.

Alongside sedentary behaviours, the method adopted in this study allowed us to estimate time in more active behaviours. Students in our sample appeared to have a very low levels of leisure time physical activity in the form of sports/ exercise averaging on 14-15 minutes weekdays and 10-12 minutes during weekends, which is much less.

In addition to targeting education in sedentary behaviour our data suggest that a major effort is needed to promote leisure time physical activity among adolescents, whether this be through sports, formal exercise or informal forms of play etc.

This study has provided useful explanatory and descriptive data on sedentary and active behavioural pattern of leisure time use of adolescent students of Ludhiana in India.

CONCLUSION

This survey has concluded that there is prevalence of sedentary lifestyle and decreased physical activity among school going adolescents. Our sample showed that there was very low level of leisure time physical activity among school going adolescents as recommended by W.H.O,[19] which states children and youth should accumulate in at least 60 minutes of physical activity per day. So, there is major need to promote leisure time physical activity among adolescents to have a better health in future, prevent obesity and other health related problems.

FUTURE SCOPE

This study has provided useful explanatory and descriptive data on sedentary and active behavioural patterns of leisure time use of the adolescents. Such data may prove useful in gaining a more detailed understanding of sedentary and active behaviours for future interventions, including better health education promotion as well as monitoring of trends .Moreover this study can be done on a larger sample size and in different schools in different areas to know the exact situation in strata.

LIMITATIONS

The survey is limited by sample size of the subjects which was restricted to 100. The survey was carried out among school going adolescents of Ludhiana, if it had been done in other schools or in a wider area, the results might have been different.

Table 2 A

0 minute / day	1 – 30 minutes/day	31- 60 minutes/day	61- 120 minutes/day	>120 minutes/day				
1	2	3	4	5				
0 MIN	15 MIN	45 MIN	90 MIN	150 MIN				
Average Time	QNO.1: I like to watch T.V., Listen music, sit and talk with family / friends.	QNO.3: I use computer ,play videogames and other sedentary hobbies (crafts, card	TOTAL Screen TIME					
WEEKDAYS	74	60	133					
WEEKENDS	83	78	161					
Average/ SD Time	QNO.1: I like to watch T.V., Listen music, sit and talk with family / friends.	QNO.2 : I spend my time in doing homework, taking tutions and readingbooks daily.	QNO.3: I use computer ,play videogames and other sedentary hobbies (crafts, card games etc) daily.	QNO.4 : I spend my time on motorized transport (bus, car, scooter).	QNO.5 : I spend my time in playing outdoor games and sports	QNO.6 : I spend time on doing other physical hobbies (gardening etc).	QNO.7: I like to do yoga and other exercises daily	QNO.8 : I spend my time on active transport (walking/cycling)
WEEKDAYS(Avg)	74	90	60	42	2	4	6	3
WEEKENDS(Avg)	83	90	78	24	6	6	0	3
WEEKDAYS(SD)	28.24	36.65	25.36	30.10	5.07	6.53	9.11	5.91
WEEKENDS(SD)	43.82	19.94	39.23	33.31	13.80	7.39	2.57	6.03

Fig 2 B

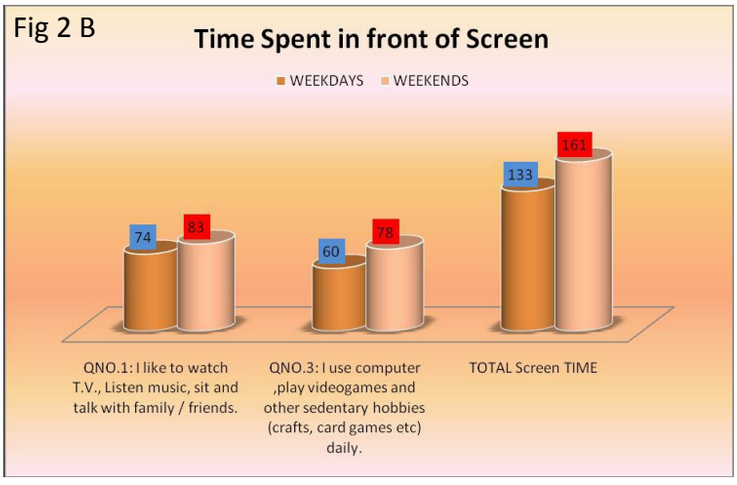


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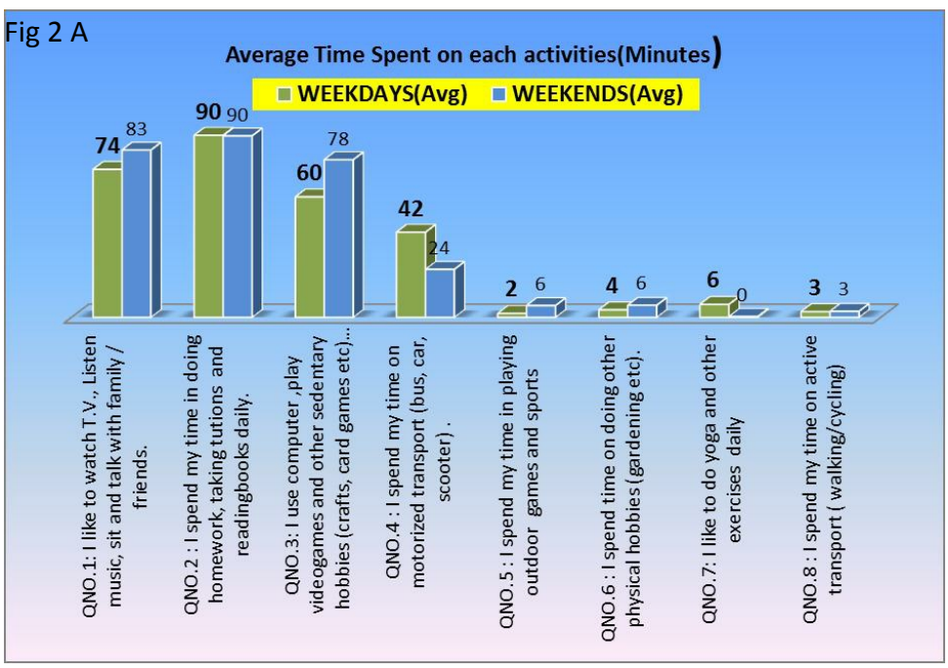
Fig 2 A



COVER STORY

Development of Blood Tests for Early Cancer Detection

By
Brian Schaefer, D.Phil. (Oxon)
CARE Biotechnologies Inc.

Abstract

CARE Biotechnologies Inc is a biotechnology company formed to initially build upon the knowledge obtained through the program of research and development of Salvestrols and CYP1B1 and extend that knowledge to development of clinical tools for the detection and monitoring of cancer. This article is intended to provide the reader with an inside look at the process of translating this understanding into clinical tools, the challenges faced and the progress that has been made.

Introduction

The development of these blood tests for early cancer detection and monitoring build upon two central bodies of research – research into the anti-cancer activities of the phytonutrients known as Salvestrols and the research into the metabolic activity of the enzyme CYP1B1. Many of you will be familiar with Salvestrols. For those that are not Salvestrols are food-based, secondary plant metabolites that have a specific functional relationship to a cytochrome p450 enzyme called CYP1B1. CYP1B1 is a unique cytochrome p450 in that it doesn't occur in healthy tissue – it only occurs in cancer cells. It is also unique in that it is a universal cancer marker occurring in every cancer tested.

What the research team discovered is that Salvestrols work as natural prodrugs. They are exceptionally stable and non-reactive compounds that bind with CYP1B1. The metabolism that results from this binding produces a metabolite that induces apoptosis in the cancer cell. This is the basic discovery of the research team in Leicester, England.

Background Research and Development

Since the work that I want to introduce builds upon the earlier research on the anticancer relationship between Salvestrols and CYP1B1 it is important to provide a brief history of this development. There are two key scientists behind this development: Prof. Gerry Potter and Prof. Dan Burke. Gerry is a medicinal chemist and cancer drug designer. Dan is an emeritus professor of pharmaceutical metabolism, and an expert in cytochrome p450 enzymes and toxicology.

Close to 20 years ago Gerry designed his first prodrug. The London based Institute of Cancer Research group that he was with, at the time, were working on prostate cancer and had a great interest in CYP17. Gerry examined CYP17 in detail, worked out its binding site for metabolic action and designed a prodrug that targeted CYP17 to perform a terminal inhibition. The drug is called abiraterone acetate, currently known by the trade name ZYTIGA®. The clinical trials of this prodrug have been exceptionally successful. The company that has taken abiraterone through clinical trials, Cougar Biotechnologies, just sold for $1billion to Johnson and Johnson on the strength of this drug. Given this drug's performance Gerry has been named cancer researcher of the year by a UK prostate cancer foundation.

Around the same time as Gerry was designing abiraterone, Dan Burke began his investigation of a new p450 CYP1B1. He found it in soft tissue sarcomas. He later found it in cancers of the breast and subsequently found it a variety of cancers including cancers of the breast, colon, lung, oesophagus, skin, lymph node, brain, and testis with no detectable presence in healthy tissue. Dan and his graduate students pioneered the research that established CYP1B1 as a universal cancer marker.

Dan and Gerry met some years after these two pieces of work. When Dan told Gerry of his exciting new enzyme Gerry immediately drew upon his experience designing abiraterone for CYP17 and started looking at how he could design a drug to target it. Within two weeks of hearing of this enzyme Gerry designed a prodrug that was metabolised by CYP1B1 to produce a metabolite that killed the cancer. Cell line results with this drug were remarkable. The drug was perfected over the next nine months, patented, sold off and the long road to clinical trials began.

During this time Gerry believed that CYP1B1 was a natural rescue mechanism and if this hypothesis was true there had to be analogues of his prodrug in food. Gerry led a search for these analogues, found quite a few of them and the subsequent research program surrounding salvestrols was born. Incidentally Gerry's prodrug has an unbelievably high selectivity for cancer cells – over the past few years natural analogues of this prodrug have been found that have vastly higher selectivity than the prodrug that Gerry designed.

Need For New Clinical Tools

As the research developed conversations about the need for better clinical tools kept arising. The difficulty was twofold.

COVER STORY

Current technology can only detect cancer once the cancer has achieved between 10^8 and 10^9 cells (if you look at the nail on your little finger, half that size is between 10^8 and 10^9 cells – roughly the size of a pea) – once cancer reaches 10^{12} cells (about a litre of cells) you're dead. By the time modern technology can tell you that you have this disease the disease has silently grown through about 75% of its life. Dan Burke wrote an excellent article on this subject titled the 'Silent Growth of Cancer and its Implications for Nutritional Protection'.

The other side of the problem is that once you are told that you have this disease the clinical tools for monitoring disease progression and treatment efficacy are poor for most of the cancers.

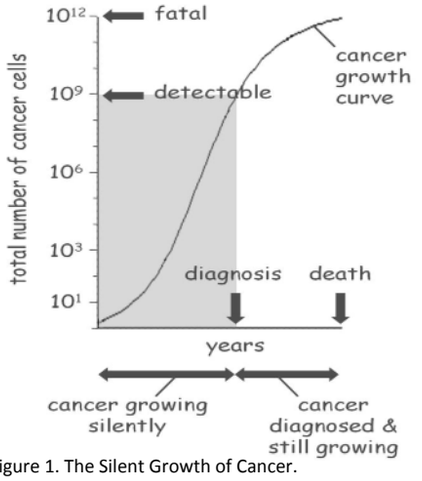


Figure 1. The Silent Growth of Cancer.
Reprinted with the kind permission of Prof. Dan Burke.

Figure 1 has implications for people that start to think about cancer prevention. They, of course, assume that they are free of the disease. They may consult a physician and take advice on cancer prevention, but this advice is likely to also assume that they are free of the disease. However, they may lie anywhere on that curve below the level of detection. If they are already up that curve preventive doses are going to slow the rate of growth but not keep the cancer from breaking through into the detectable range.

Figure 1 also has implications for people that have journeyed through this disease to the point that their physician has told them that they are 'all clear'. This statement may simply mean that their disease is again below the level of detection. In the absence of ongoing treatment and/or lifestyle change their cancer may break through into the detectable range over the next few years.

All in all it amounts to a pretty miserable picture.

It would be ideal to equip clinicians with a simple blood test that could be used to screen for any of the cancers, with a sensitivity that could pick up the presence of the disease long before it has reached 10^8 and 10^9 cells. Think about how much easier it would be for them to assist these people back to good health. It would also be ideal if a simple blood test could be used to monitor any of the cancers with a level of accuracy that would readily tell if a treatment is working or not and whether a dose is high enough to be effective? A blood test that is as applicable and accurate with pancreatic cancer as it is with breast cancer – a blood test that is as applicable and accurate with adrenal cancer as it is with prostate cancer. Tools such as these could make life a lot easier for clinicians and patients alike. Tools such as these would provide additional data that would help to advance our understanding of how best to prevent and treat the various cancers and how best to keep them in remission.

Development of Clinical Tools.

The need for new clinical tools is obvious. One of the enormous implications of the prior work of Profs. Potter and Burke is that it sets the stage for the realisation of blood tests such as those that I just described.

To embark on this work we looked at what we had to work with. We had great expertise on CYP enzymes, we had great expertise on secondary plant metabolites and their metabolism by CYP enzymes. Specifically, we had CYP1B1, a universal cancer marker and salvestrols, natural prodrugs, which in this context amounts to things to look for in the bodily fluids of cancer sufferers. Given the salvestrol – CYP1B1 mechanism we knew that there would be things that we could look for that would tell us about the presence and state of this disease. Basically we could use our understanding of this metabolic relationship to report back to us on the disease itself.

We took the decision to utilise this knowhow and develop clinical tools for the early detection of cancer, and treatment efficacy. One thing that we have learned so far on this project is that it is a really good idea to have people on your research team that don't realise 'that it can't be done'.

In considering the problem we decided that we had one of two directions to take. The obvious first route was to develop a method for detecting and measuring the presence of CYP1B1 itself. Since CYP1B1 is an intrinsic component of cancer cells its detection and measurement would provide a direct measure of the disease itself. The second and much less obvious approach was to develop a method for detecting and measuring the metabolic output of CYP1B1. If we could find a strong metabolic output of CYP1B1, detect and measure it we would have another direct measure of the output of this disease and by extension the disease itself. So we decided to pursue both – two universal tests for cancer.

The Proteomic Approach:
In pursuing the detection and measurement of CYP1B1 itself we knew that the job would be made much easier if we had antibody as this would help us to isolate what we were looking for from all of the background material in human blood, urine or tissue. We wanted an antibody to an amino acid string that was 100% specific to CYP1B1, covering the wild form and the major polymorphs and not found in any bacteria and one that didn't have major cleavage sites running through the middle of it. These criteria ruled out all of the antibodies that are currently available for CYP1B1. We performed an exhaustive search and identified a set of peptides that met our criteria and embarked on raising antibodies.

CYP1B1 is a very difficult enzyme to raise antibodies for that have a strong affinity for the peptide of interest because CYP1B1 is present in so many life forms in identical form or near identical form to

that found in humans. However, we did manage to raise an antibody to a specific CYP1B1 peptide and worked on affinity enhancement until we had something useable.

Our first notion was to see if we could detect and measure CYP1B1 in human tumour samples. Seemed like a good idea at the time – where else are we going to find CYP1B1 in abundance?

We spent about a year working on sample preparation methods and testing samples using some of the world's most sophisticated mass spectrometry equipment. We spiked the tissue matrix with CYP1B1 from recombinant sources and managed to recover the recombinant material but never managed to detect the native CYP1B1. This caused us some considerable concern because the wisdom of the day dictated that if we couldn't manage to detect and measure CYP1B1 in tumour samples, where it would be plentiful, we would never be able to detect and measure it in blood or urine. However, given that we were able to detect and measure the recombinant CYP from the tumour matrix we knew that we had a sample preparation and extraction problem – we were either not freeing the enzyme from the surrounding material or we were destroying the enzyme with our preparation method.

In light of this we decided to abandon our search for CYP1B1 in tissue and focus on detecting it in blood. This decision flew in the face of conventional wisdom but our thought was that if we were ever going to have a viable diagnostic and monitoring tool it had to work on blood or urine samples so if we were going to pound our heads against a wall it might as well be the wall we needed to get to. This new strategy isn't really as crazy as it initially sounds. When working with blood you don't need some of the sample prep steps that you would use with tissue because you don't have as much intact material to deal with – you are already working with fragments.

So we embarked on trying to find our CYP1B1 peptide in blood and ended up with the same results as we found for tissue! We spiked recombinant CYP1B1 into blood and managed to recover it but were unable to recover native CYP1B1 amidst a chorus of 'I told you so' until one member of the team came up with the bright idea of starting with more blood! We increased the initial sample size and detected and measured our native peptide.

Results

The naturally present CYP1B1 peptide was successfully detected using antibody-affinity capture in both 20 µl and 200 µl digests of cancer patient plasma. The amount of natural CYP1B1 in this sample can be estimated to be ~200 amol/µl of plasma. Upon achieving this result we had an opportunity to replicate with 5 additional samples.

Sample	Amount of CYP 1B1 (amol/-l of plasma)
2ml tube labelled 22/04/2009	12.5
2ml tube labelled 23/04/2009	2
2ml tube labelled 24/04/2009	9.4
2ml tube labelled 25/04/2009	9.2
1.5ml brown tube non-labelled	4.9

Figure 2. Initial set of results - plasma levels of CYP1B1.

Lower levels of peptide were found in these samples with the amount of natural CYP1B1 ranging from 2 to 12.5 amol/ µl of plasma. (MRM analysis was performed by injecting 15 µl on an AB/MDS Sciex 4000 triple quadrupole mass spectrometer equipped with a Nanoflow Eksigent NanoLC-1Dplus HPLC, 5 x 0.3 mm C18 Pepmap (5 µm particles) trap column, and a 75 µm x 150 mm Magic C18AQ (5 µm particles, 100 Å pore size) analytical column using 30 min. analyses.)

Since achieving this early result we have refined our methods and analysed plasma represented a small range of cancers. We have also analysed plasma from a proteomic standard to serve as our baseline. The proteomic standard represents pooled plasma from healthy volunteers. We have detected CYP1B1 in this proteomic standard at minute levels that can only represent the CYP1B1 found in those tiny number of pre-cancerous or cancerous cells that are being killed off in healthy individuals on a daily basis. With plasma from cancer sufferers our best results to date have been found with lung cancer samples. When comparing CYP1B1 levels to those in the proteomic standard the levels in the lung cancer samples were vastly higher (between 92 and 6291 times that found in the standard).

With these results we performed some calculations to determine where these results would fit into the growth curve of the 'Silent Growth of Cancer' and concluded that these results would provide detection of disease around 5.7 years earlier than existing technology. In terms of the prognosis for lung cancer this 5.7 year difference could be the difference between life and death.

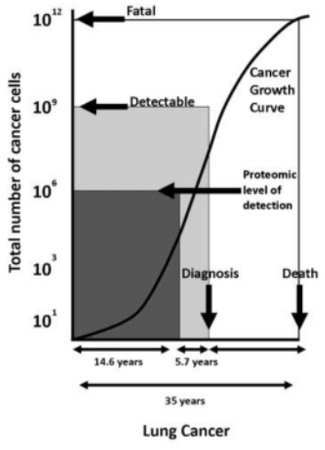


Figure 3. Level of detection of CYP1B1 in lung cancer samples.

Summary

We now have a sample preparation method and an antibody (an assay) that is able to directly detect and measure cancer though detection of CYP1B1 in plasma. When we find our peptide in your blood with this assay you have cancer – there are no false positives – you have cancer.

We now have a variety of method enhancement experiments, stability experiments, validation experiments and method transfer experiments to conduct but we know that CYP1B1 is present in the blood, we can find it and we can measure it.

What I really like about this approach is that it will be simple and convenient for the person getting tested. Simply put out your arm for a sample collection like any other blood test. What I also like about this approach is that it is a direct detection and measurement of the cancer itself and it is as applicable to pancreatic cancer as it is to breast cancer – it is applicable to all the cancers. The other thing that I like about this test is that we are operating at an exceptionally high level of sensitivity and we have good reason to believe that

we can increase the level of sensitivity from here.

The Metabolite Approach

We know the various substrates of CYP1B1, that is, we know what it metabolises and in particular we know a lot about the salvestrols that it metabolises. So what happens when we ingest salvestrols?

In our food salvestrols come in two forms: as a glycoside and as an aglycone – in food about 80% as glycosides and 20% aglycones – in supplements 100% aglycones. When we ingest the glycoside the plant sugar is cleaved off and replaced with a human sugar. When we ingest the aglycone a human sugar is attached. This of course assumes that everything is working properly to perform this function. The new glycoside is then transported and upon reaching cancer cells the human sugar is cleaved off leaving the aglycone at the cancer site. This step is performed by Beta Glucoronidase. The aglycone then binds with CYP1B1 and is metabolized. The metabolite induces apoptosis spilling the contents of the cancer cell, including CYP1B1 peptides and metabolites into the surrounding space.

What all this means for blood test development is that the interaction of CYP1B1 and salvestrols provides us with a variety of measurable aspects of this process that can provide us with insights into the presence of disease as certain of these aspects can only be present if the disease is present and metabolism has taken place.

We initially went through our list of salvestrols looking for metabolites that were abundantly produced through CYP1B1 metabolism and not found in a typical diet. From a candidate list one metabolite was chosen.

We looked to see if we could find the aglycone in blood and urine – initially using predicted structures and then using synthesized standards we were able to reliably detect and measure the aglycone in both blood and urine. We then performed a pharmacokinetic study using healthy volunteers to determine when salvestrols reach peak concentration in the blood – three hours after ingestion. We identified the aglycone spike resulting from salvestrol using hplc. Prior to hplc analysis the samples were prepared and Beta Glucoronidase was used to remove the sugar from the glycoside.

Following this we decided to take a look and see if we could find a difference between healthy volunteers and those with advanced cancers. We administered 1 gram of a specific salvestrol to each individual, waited 3 hours and drew their blood. We also had each individual do a 24 hour urine collection. As expected, with healthy volunteers we found no metabolite – we simply recovered the substrate in blood and urine. With diseased volunteers the situation was very different. We found a very clear spike on the hplc where we predicted that the metabolite should come off the column. Some of these individuals had very advanced disease and with these individuals we found absolutely no aglycone and no glycoside – just metabolite. When we analysed their urine we also found no aglycone. The entire gram of substrate seemed to have been used up. With other cancer patients we found small amounts of aglycone along with large metabolite spikes. What this tells us is that the ratio of metabolite to aglycone may be of much greater clinical value than the metabolite alone – time will tell. We performed these tests with individuals representing a fairly broad array of common cancers: breast, stomach, kidney, prostate, etc., and an array of stages of cancer but skewed towards more advanced cancers. Metabolite spikes were found for all as one would expect given that we are looking at the metabolic output of a universal cancer marker.

Summary

We currently have a sample preparation method that allows us to detect the aglycone and the metabolite in blood or urine using hplc. We find clear separations between the outputs obtained from healthy volunteers as compared to diseased volunteers. Like the proteomic approach when we find this metabolite in your blood you have cancer.

What I really like about this approach is that it uses natural products as diagnostics. We are getting the metabolism of a natural product to report on the presence and state of disease. Another nice feature of this approach is that we can build the signal by the amount of substrate that we administer. An additional benefit of this approach is that it not only tells us that CYP1B1 is present, that is that cancer is present, it can tell us that the enzyme is functioning fine.

There is also great scope for improving this assay. We have been drawing blood at 3 hours, the time of peak concentration for the substrate. Conduct of a small pharmacokinetic study to determine peak concentration of the metabolite will benefit this test greatly. Once we are able to draw blood coincident with peak concentration of the metabolite we will be able to pick up the presence of cancer much earlier. Like the proteomic test the metabolite test is universally applicable.

Discussion

A present we have two different assays for detecting and measuring the presence and amount of cancer. Both operate independent of any apriori notions about type of cancer that may be present. The huge strength of these approaches is that they can be used with all cancers – they are two universal cancer tests that can ultimately be used for diagnosis and monitoring across all of the cancers. The downside of this is that we will need to validate both approaches on each and every cancer.

Up untill now everyone on the team has had their pet blood test – either the metabolite test or the proteomic test. However, from the onset there have been good arguments for seeing both of these approaches through to completion because they provide differing set of strengths and weaknesses and when we combine them we can potentially provide much more clinical assistance than we could with results from either one.

For example, let's say that we have two 36 year old females, very similar in family history, medical history, etc., and both have a 2 cm cancerous lump in one of their breasts. Their physician decides to run the metabolite test. With one of the women a large metabolite spike is found with no aglycone and no glycoside. With the other woman we find a medium sized spike of metabolite and small spikes of aglycone and glycoside. What is going on? With just the metabolite test we might conclude that woman number 1 has fully

functioning CYP1B1 that is making full use of the substrate while woman number 2 may have competing substrates in her body that are inhibiting the functioning of CYP1B1 – for example she may have been using some household paints that contain chemical anti-fungal agents or perhaps she had the furnace duct work recently cleaned and the cleaners used chemical anti-fungal agents to retard any fungal build up or perhaps she takes daily walks along the perimeter of a golf course that uses a great deal of anti-fungal spraying. We may also conclude that woman number 1 may have an additional undetected tumour mass. If we now run the proteomic test we can help to determine what in fact is going on for these two women. Let's say that we run the proteomic test and find a larger spike of peptide for woman number 1 than woman number 2. This result would tell us that there may be no difference at all between the functioning of CYP1B1 for these two women but rather confirm that woman number 1 has another, undetected tumour mass and this is accounting for the higher results. The attending clinician can then embark on a search for the whereabouts of this second tumour mass.

Current Directions

We want to keep pushing the limits of detection until we are picking this disease up at disease onset.

We want to be able to pick this disease up at points where simple dietary and lifestyle change can turn the disease around. That is one of our goals.

We also want to push the sensitivity of these tests to the point where we can tell whether or not a treatment is working within 24 or 48 hours after commencing treatment. This will be exceptionally beneficial to people pursuing conventional chemotherapy as it could save them from weeks of toxic exposure.

We also want to be able to utilise these tests to individualise treatment plans for cancer sufferers.

We want to push the sensitivity of these tests to the point that we can pick up disease recurrence at a point where dietary and lifestyle change can turn the disease around.

Along the way to reaching these goals we are able to add value to those volunteers that contribute their blood through advising their physicians of the results.

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About the author:

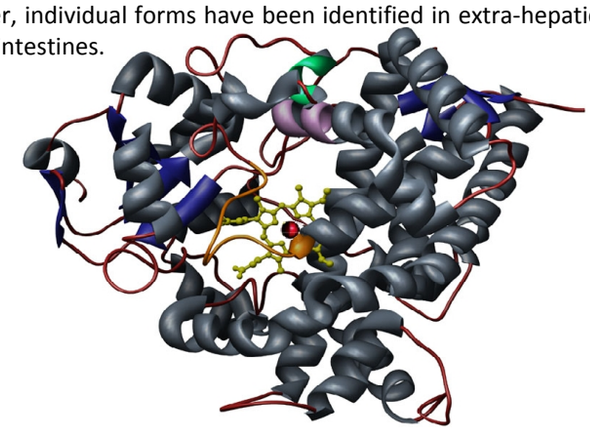
The author was educated in Victoria, B.C., Canada and Oxford, England, obtained a B.Sc., and M.Sc., degree from the University of Victoria and a Doctor of Philosophy (D.Phil.) degree from Oxford University in England (Wolfson College). After these studies were completed he chose to return to Canada. After two years as a research fellow in Ottawa he returned to Victoria where he currently lives with his wife and his two children. A fondness for England continues and he returns to England on a regular basis. He has published and lectured on a broad array of topics including psychometrics, pattern recognition, visual perception, knowledge acquisition, artificial intelligence, laboratory medicine and cancer research.

The cover story of our first issue is an in depth explanation of CYP1B1, If you have missed it then it is available on our website as a download. The following is an extract from it:

Cytochrome P450 enzymes (abbreviated as CYPs and pronounced 'sips') belong to a superfamily of proteins which contain iron at their core and often referred to as 'haemoproteins'. These CYPs are a large and diverse group of enzymes that use a variety of large and small molecules as substrates in enzymatic reactions. Being enzymes they are natural biological enabling devices which change structures of chemicals in the body without themselves becoming changed. CYP enzymes have a long history and originated 3.5 billion years ago. Their name is a composite one derived from having a cellular location(cyto), a spectrophotometric characteristic (chrome) and a unique optical absorbance peak at or near 450nm.

CYP enzymes are ubiquitous and found in all life forms from animals, plants and fungi. More than 11,500 distinct Cytochrome P450 enzymes are known to exist. In humans there are 57 different CYP proteins wich are all bound to membranes of the endoplasmic reticulum or to the inner membrane of the mitochondria and as such have distinct cell-specific, tissue-specific and developmental-stage-specific characteristics. The majority of CYPs are to be found in the liver, however, individual forms have been identified in extra-hepatic tissues such as lung, kidney and intestines.

Right: CYP1B1



CLINICAL CORRESPONDENCE

Case Study:

Low grade papillary urothelial carcinoma, kidney cancer and pancreatic cancer.

B.A. Schaefer, D.Phil.(Oxon)

A 61-year-old male presented to his doctor with macroscopic haematuria, swollen ankles and fungal infections on his nails (Onychomycosis with concurrent Paronychia). He was a non-smoker with a history of good health outside of developing septicaemia following a knee operation eighteen years earlier. His bleeding persisted for two days and his doctor, although suspecting an infection and prescribing an antibiotic, referred him for an ultrasound. Two weeks later an ultrasound revealed an abnormal thickening near the left vesico-urethral junction. He was referred for flexible cystoscopy.

After a period of seven months flexible cystoscopy revealed a small papillary tumour near the bladder neck close to the left vesico-urethral junction. An enlarged prostate was also noted. He was referred for surgery.

Two months later a transurethral resection of bladder tumour (TURBT) was performed. Surgery revealed a 2cm papillary growth at the bladder neck and overlying left ureteric orifice. No obvious disease was found in the ureter. A single dose of epirubicin was installed. Paracetamol (Panadol tablets) 500mg, twice daily along with tramadol hydrochloride, 50mg capsules, 1-2 daily, were given for pain relief but declined. Sodium citrotartrate (Ural) granules, 1 sachet daily, was also prescribed. During surgery it was discovered that the tumour also involved part of the urethra and lesions were also found on a kidney. The surgeon felt that the cancer had been removed from the bladder and urethra but was concerned about the kidney lesions and unsure about whether the bladder cancer would return. The swelling in his ankles was quickly resolved post-surgery.

The histology report indicated that a grey/brown tumour, 20mm x 17mm in aggregate was received post-surgically revealing a low grade papillary urothelial carcinoma with no invasion present. He was referred to an oncologist. The oncologist indicated that the tumour had been a fast growing tumour that was quite rare. It was larger than they had anticipated. Flexible cystoscopy and *magnetic resonance imaging (MRI) were scheduled as part of a post-surgery, follow-up.*

Seven months following surgery flexible cystoscopy indicated a tumour on the pancreas. An MRI was performed six weeks later revealing a 40mm tumour on the pancreas. The oncologist informed the patient that he had a life expectancy between 10 and 20 weeks. The patient became overcome with fear as he was very familiar with the prognosis for pancreatic cancer due to his prior involvement with pancreatic cancer research fundraising.

Coincidently a former colleague had been in touch and upon hearing of the cancer diagnosis suggested that he supplement his diet with Salvestrols. On three separate occasions his colleague made this recommendation and his suggestion was declined. Two weeks following the diagnosis of pancreatic cancer he agreed and commenced daily Salvestrol supplementation comprising two Salvestrol Platinum capsules for a total daily intake of 4000 Salvestrol points. This was added to his daily regimen of 500mg of vitamin C, a fish oil capsule and an evening primrose capsule. No other changes to diet and lifestyle were made at this time although there was a modest move towards more organically grown foods. Daily exercise had always formed part of his routine.

Twelve weeks following his MRI confirmation of pancreatic cancer and eight weeks following the onset of Salvestrol supplementation flexible cystoscopy indicated slight shrinkage of the pancreatic tumour. Follow up MRI confirmed the shrinkage and revealed concurrent shrinkage of the kidney tumour. Salvestrol supplementation was maintained along with his usual daily supplements. No further changes were made to diet or lifestyle.

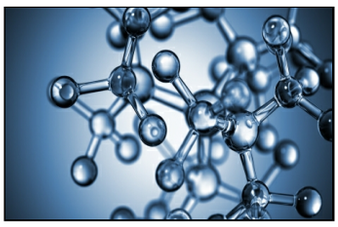
Four months later and six months after starting Salvestrol supplementation a further flexible cystoscopy found no evidence of pancreatic or kidney cancer. Given this surprising result an MRI was scheduled. The MRI confirmed the finding of the flexible cystoscopy. Due to the highly unusual finding a second MRI was performed which also confirmed the findings of the flexible cystoscopy. An astonished oncologist told him that he was cancer free and that the kidney had healed very well. A follow up flexible cystoscopy and MRI were scheduled in a year's time.

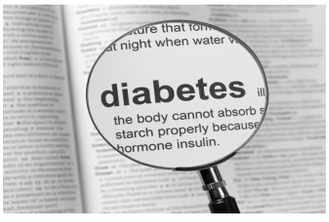
During the intervening period he has decreased his daily Salvestrol supplementation to one Salvestrol Platinum capsule per day for a daily total of 2000 Salvestrol points. He has commenced a daily, one hour walk and moved his diet to include organically grown foods when ever practical to do so. He is feeling great and enjoying good health. Twelve months following his cancer free status he reports that he feels ten years younger and his recurrent problems with nail fungus (Onychomycosis with concurrent Paronychia) have also been resolved. Salvestrol cream was applied for this purpose.

As the date of his one year follow up for flexible cystoscopy and MRI approached, Salvestrol supplementation was increased to two capsules comprising a total daily intake of 4000 Salvestrol points and then three capsules for a daily intake of 6,000 Salvestrol points. The one year flexible cystoscopy found no evidence of bladder cancer, pancreatic cancer or kidney cancer. His cancer free status was confirmed and it was felt that there was no need for an MRI. He will be tested again in a year's time.

diabetescan

ADVERT





Today 346 million people worldwide suffer from diabetes, a chronic disease when the body does not produce enough insulin, or cannot effectively use the insulin it makes. With 1 in every 20 people suffering from diabetes population screening and cost effective monitoring becomes essential.

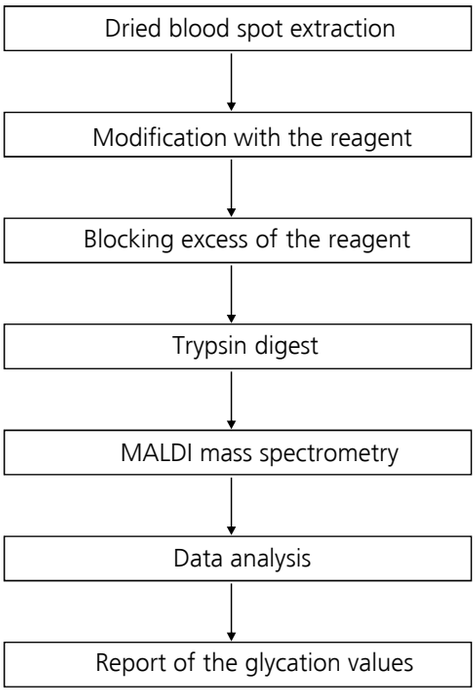
Diabetes: A modern disease

Insulin is a hormone in the body that regulates blood sugar. Over time, raised blood sugar (hyperglycaemia) can lead to serious damage of the body's systems, particularly the blood vessels and nerves.

In the long term, diabetes can damage the heart, blood vessels, eyes, kidneys, and nerves. It increases the risk of heart disease and stroke, blindness and death.

There are many people who have diabetes and remain undiagnosed until complications arise.

Diabetes and its complications have a significant economic impact on individuals, families, health systems and countries.



A new diagnostic test for diabetes

Scientists have recently discovered a new and inexpensive way of testing for diabetes.

The test is based on a known diagnostic marker, called glycated hemoglobin (HbA1C).

In the life of a red blood cell, glucose molecules react with hemoglobin to form glycosolated hemoglobin. In individuals with high blood sugar due to poorly controlled diabetes, these molecules are more prevalent.

The test is an accurate, low cost, robust method of diagnosing diabetes. Furthermore, the test can detect these molecules in a spot of dried blood, making this method suitable for mass screening a population.

Diabetescan

- ·Diabetescan delivers a robust method of diagnosing diabetes
- ·Diabetescan delivers accuracy capable of diagnosing diabetes from a spot of dried blood
- ·Diabetescan delivers high throughput for rapid, cost-effective results on a nationwide scale

CARE
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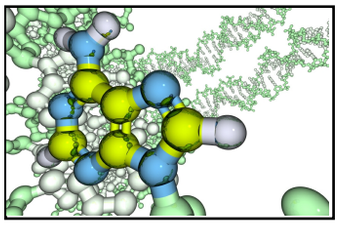
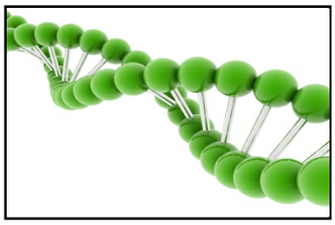
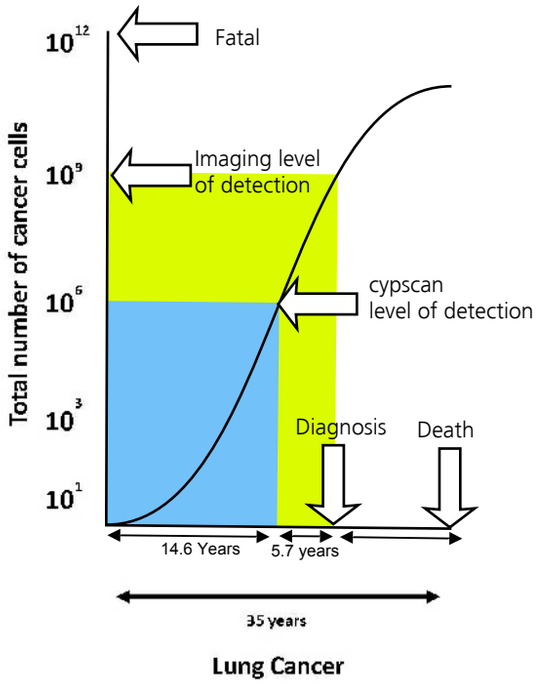



These days everyone knows how prevalent cancer is. Many of us have lost loved ones, work colleagues or friends to the disease, or have seen others battle through it. Still the figures are shocking. The Cancer Research Agency in the UK states that an incredible one in three will develop some form of cancer in their lifetime.

Early detection is key

One of the biggest problems facing physicians in the treatment of cancer is early detection. Although there are some screening tests,cervical smears, mammograms. All of which are imperfect tests as they are dependent on the skills of the pathologist looking at the sample many people are left to discover they have cancer when they experience unusual symptoms or discover a lump. Unfortunately, at this point the cancer can already be quite far advanced and may have even spread to other organs, making treatment much less effective.



A new screening method that will save lives

A unique enzyme called CYP1B1 that is produced by cancer cells. The importance of this enzyme has lead to a revolutionary new blood test that has been developed to detect any cancer at very early stages of its development if cancer is found early enough, simple changes to diet and lifestyle could be enough to prevent the progression of the disease. This is a huge step forward in cancer diagnosis and treatment. It is estimated that one third of all cancers could be cured, using current conventional methods, if detected early and treated appropriately. This would mean that this new cancer test could save millions of lives worldwide every year.

Cypscan

- Cypscan delivers early detection of all cancers (detection of lung cancer 5.7 years earlier than medical imaging can deliver)
- Cypscan delivers cost-effective monitoring of all cancers
- Cypscan delivers high throughput for rapid, cost-effective results on a nationwide scale

CARE BIOTECHNOLOGIES

CARE BIOTECHNOLOGIES

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Website: www.carebiotech.com

Care Bio-technologies has identified a unique cancer specific signature enzyme which enables early diagnosis of cancer to occur so ensuring that meaningful interventions can be made well before symptoms-which are usually the first indications of disease-occur.

Our advances to facilitate this early diagnosis of the disease will save lives, significantly improve patient outcomes, and enable clinicians to engage in a meaningful process of prevention, all at a substantially enhanced level compared to that which even the most advanced present day technology can provide.

Our technology will identify the significant presence of cancer cells without the need for any a priori idea of where in the body the cancer is located. In so doing it provides a relatively inexpensive first stage indicator as to which individuals should go for more expensive scanning and biopsy cancer tests, and it also enables clinicians to assess the progression of disease and provide an indication of the success or otherwise of any selected management programme.

Significant investment and directed research has already taken place to establish the foundations for this new patented technology and as a result we have identified a number of different approaches which have the potential to be executed using current commonly available analytical techniques used in commercial laboratories.

Our two lead researchers have a wealth of experience in cancer discovery behind them and these advances represent a new approach to the conundrum of how to assess and evaluate the presence of cancer in an individual. Our approach uses simple analytical assessments of blood or urine specimens and in the future, possibly breath, without the need for invasive methods, biopsies or highly expensive and complex in-vivo monitoring devices.

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