Research into

Improving Sperm Quality

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ABOUT THE AUTHOR

In this eBook I share an explanation of life that is obvious – hidden in plain view. As a contribution to the mysteries of becoming pregnant one needs just to go back to what always worked.

A simple life, and more happy sex.

As someone who came from a time when becoming pregnant was as simple as unprotected sex – once - my position is 'of course you will get pregnant.' As it has always been, in all other times of human history, sex equals babies.

I have simplified this as much as possible. Simple. Natural. Solutions. These three words solidify my work in restoring health, wellness and vitality to everyday life. You may wonder - it can't be that simple surely? Used to be. Often it is.

I have been working with couples to assist natural fertility and great pregnancies for over 35 years. As a mother, I cannot imagine a life without children – or without the focus on the future that children bring. They give such joy and completeness to a life. Follow nature. Of course you can make babies.

As a natural healer I gravitated to what may have appeared to others to be absolutely 'hopeless'. When I started working in the field, to have blocked tubes (male or female) or to have a poor sperm count, or 'unexplained infertility', couples had no option but to see someone as myself. Working with enhancing the body's ability to heal itself and to restore natural normal functioning works.

As a problem-solving health care professional in the past decades, I have been able to bring the very different focus of Chinese medicine into play for others to see how easy life changes affect health and baby outcomes. Fusing natural healing information from both - natural healing traditions, and using the Chinese medical model, allows the motivated person to take full responsibility for themselves.

As a naturopath, I help you see how making lifestyle changes, rather than letting life destructive habits co-exist with following in fear and hope, expensive medical ideas or other paid-for solutions. Just being healthier is usually all that is needed to become a dad. Staying where you were when you created your not so good sperm issues issue is not the answer.

I aim to remove whatever is blocking normal. Life naturally brings forth babies, as this is what we are here for. No need to check if the baby is growing well, as if baby is started with the best ingredients, nature takes its course perfectly.

Your life changes create better babies and an easier life for all – especially the baby who has to live in the body that you make, forever.

DISCLAIMER

The information in this eBook is intended as a general guide only and is not a substitute for the individualized health care. It has been written to fill a gap that may be missing in your life – why things have happened and how you can help yourself.

In most of your life, what you put in is obvious by the results. As you live with the consequences of the actions you choose – often decades before it is obvious what you have set in motion.

Especially when making babies - as you make them their own ovaries and sperm generating cells - your grandchildren's beginnings – are also being made.

I recommend that you really think through any options given from your health care practitioner, midwife, or doctor. Any health concerns you have for yourself or your family in general health and wellbeing, especially in all matters relating to pregnancy, birth and in parenting, need investigating. . .

Second, third and sometimes more opinions may be needed.

I suggest always following Nature first.

That said, there is also the issue of readability:

Due to the urgency I feel in getting my life work 'out there', and with the lack of my personal resources, it is very possible that a critical eye may find typos, links that are no longer working and grammatical and writing errors. Perfection may come later.

My focus is on getting information to you the reader, so that you have more choices, especially in the areas that presently may seem hopeless.

FOREWORD

After many decades of assisting couples in their journey to become families, I am still amazed at how pieces of the jigsaw are not all put on the table when you are trying to complete your puzzle. By anyone. You may find that just starting from the actual beginning – your own state of health - is all that is necessary to improve your sperm.

Making babies has become very big business. WHY sex is not making babies as it used to, is not being asked. There is now an epidemic of unhealthy people believing that it is just a matter of spending money to essentially buy their perfect pregnancy.

Assisted reproductive seems to rely on a balance of protocols and sometimes results with occasional babies being made. Often with no answers as to why it did not work – why the apparently great looking sperm was dud – or that the amazing Grade 1 embryo fizzled . . All without consulting nature: why not naturally? Is there something that needs addressing first?

To try to help you here, some of the medical specialists are now venturing outside their own scope of practice – which is surgery and pharmaceutical prescribing - into seemingly knowing what to 'prescribe' as supplements and lifestyle changes. BUT – working holistically for you is not their training. It may be a matter of undoing what should not be there first, and then returning you to simple living. Unlikely adding a few vitamins to the mix – the ones research have been found to 'help' – is all.

Without a substantial mind set shift, supplements alone may not make a perfect baby. I suggest here that you become more conscious. If this project were a new house, more effort would be put into ensuring you get all that you want. Not as with pregnancies begun - hopeful scanning later, but real effort to make sure FIRST. You can not easily redo foundations – you can only make that baby once.

The body heals itself. This is not taught in medically orientated courses – of any description. Health restoration is about personal effort and changing habits. Not someone doing something to you, or selling you a product.

Thinking for yourself

Extrapolation is needed, as often there are just animal studies to look at. If something worked for rats - why would you not do it yourself? As an example, decades ago I heard that supplementing pregnant rats' feed with Vit E stopped them reabsorbing their young (this happens also in early human pregnancies - especially the case of 'the disappearing twin'). Now there are studies done to support this.

This means taking Vit E before and during pregnancy is a protective influence. Of course the fat soluble vitamins such as E do need you to have enough dietary fat and Vit D on board. In times past all were living outside, not only inside as we do now. This means that getting back to basics is very important. Look to increasing your Vit D levels to the top of the range apparently needed according to blood test (The RDA is a guess).

Research and what it means

Life is not linear. If you want to be a parent of a spectacularly healthy child or children, it may mean that you do all that you can to raise the chances - even if it has just a 1% likelihood of making a difference. Very often nutritional deficiencies are at the root of what is happening. Formal research is about ONE variable. Life isn't.

All natural therapists have known that taking fish oil all through a pregnancy protects against early labour, and termination of the pregnancy before baby is ready to live independently. Now there are studies also to support this.

Using supplements and changing your life habits will improve your health. Safer, more effective and cheaper than using artificial reproductive services to forcibly make your baby. If you don't need the extra nutrients provided, your body will either store or expel them. It is likely that there is repair that can be done and your body will just be easier to live in. This is a very different outcome than using the many chemicals that are NOT food substances and that were never intended for human consumption.

Differences between farming and medicine

Western medicine has not grasped an agricultural truth - good ingredients need to be in the soil. Good seed stock to make great plants. Breeding stock has to be the best of what is available; not the least fertile, with external help. Please look to epigenetics - great growing conditions are needed for optimal gene expression.

Making your babies may have to start with unloading the excess toxins stored within you. This ensures you do not bequeath your children and grandchildren the residues that you may currently have within you. These are a consequence of breathing in the air we are surrounded by: through drinking the water, and by eating the food that has been grown in the waste dump this small green spaceship we all live on together.

Although medicine is not looking at the tragic consequences of modern life, detoxing can only help your health and thus your fertility and the quality of your children. Just a simple issue as avoiding all plastics may help. Reading Slow Death by Rubber Duck and 'Toxin, Toxout' would be yet another wake up call. The information as to why sperm are struggling is out there. What to do about it is also available below.

If medicine has no answers

Think for yourself. Western medicine is new – only about 150 years old - and it keeps on reinventing itself. Questions are not being asked. Knowing why even cancer or endometriosis happens AGAIN is beyond the best thinkers in conventional medicine. Or why the rate of autism continues to alarmingly increase. Even why so many new mums are not breastfeeding easily – as mammals that is what we are breed to do.

Something is 'breaking' normal. Not looking at all possibilities is not optimum, whilst you wait patiently for IVF to fix your problem – do it yourself - start by improving your sperm, and thus your chances of a better baby. Answers to your questions as to why miscarriages and stillbirths and reproductive mishaps keep happening are not to be found if the right places are not looked into.

INTRODUCTION

This eBook can to be read in conjunction with the Men's Vitality Package, and also the Helping Mother Nature – a Fertility Guide eBook. Supercharge Your Sperm is part of the text found in this one. It is also available as standalone eBook. If you want to start looking yourself to see how I came to these conclusions - please read within.

You may wonder why the medical assisted reproductive specialists seem to not know what you read within these pages. You may also know that after having many reproductive mishaps – often after years of doing everything 'right' – that there are no answers as to why there have been only miscarriages, no satisfactory embryo to transplant or even no fertilisation to show for your efforts. You may ask why no one suggested that there were very simple effective things you could do in your everyday life to alter your chance of being a dad.

I cannot answer why others have not taken the short amount of time it takes to start the Internet search. This information is hidden in plain view. It is not a secret that changing your ways will change you sperm. Change the state of your health, and you will make better sperm. This may be news to you. All farmers do operate from here.

But - conventional modern medicine is focused on disease management.

Even if you have been told it is impossible for you to become a father naturally, or at all – there may well be hope as many have tried these simple additions in their lives - and gotten rid of the sperm destroying habits – and are now very happily fathers.

What to expect within;

Chapter one outlines some of the research that is easily found when you search online for reactive oxidative stress - one of the leading issues in make infertility that you may never have heard of. This is primarily as YOU have to make changes to your life to change your sperm health. No one makes money on this deal. You do however life, your chances rise substantially of having a natural conception and a perfect baby if you do so.

Chapter two introduces the idea that the body works as a whole and that the various supplements you may think to take need to be in the context of a well functioning body/mind.

Chapter three explores the Chinese medical rationale behind looking after yourself so your gut works optimally – the sperm made will be far better quality should you take this advice.

Chapter four, five outline the digestive troubles that may await you should you miss following sound advice and chapter five offers simple solutions to these.

How to put it all together - Chapter six – food and supplements

You may just need to let go the limitations, and start living more simply, naturally.

Possibly then look through Chapter one again to see what this means to your life.

1 - STUDIES OF IMPROVING SPERM QUALITY

There are many more papers that can be sourced – just search online for 'ROS (reactive oxidative stress) and male infertility'. You will find that antioxidants work to undo this. Lifestyle changes and cleaner living mean less ROS happens.

Clinical Relevance of Oxidative Stress and Sperm Chromatin Damage in Male Infertility: An Evidence Based Analysis

Marcello Cocuzza, Suresh C. Sikka, Kelly S. Athayde, Ashok Agarwal

http://www.brazjurol.com.br/september_october_2007/Agarwal_ing_603_621.pdf

ABSTRACT
Oxidative stress (OS) in the reproductive tract is now a real entity and concern due to the potential harmful effects of high levels of reactive oxygen species (ROS) on sperm number, motility, quality, and function including damage to sperm nuclear DNA. Evaluation of OS related damage to non-functional sperm is highly relevant as intracytoplasmic sperm injection (ICSI) technique, an effective therapy for severe male factor infertility, bypasses the majority of reproductive tract deficiencies.

Despite the controversial findings in the existing literature, there is now enough evidence to show that sperm DNA damage is detrimental to reproductive outcomes. In addition, spermatozoa of infertile men are suggested to carry more DNA damage than do the spermatozoa from fertile men. Besides impairment of fertility such damage is likely to increase the transmission of genetic diseases during the assisted reproductive procedures. Standardization of protocols to assess reactive oxygen species and DNA damage is very important in introducing these tests in such clinical practice. Thus evaluation of seminal ROS levels and extent of sperm DNA damage especially in an infertile male may help develop new therapeutic strategies and improve success of assisted reproductive techniques (ART).

Mechanisms of male infertility: role of antioxidants.

Sheweita SA, Tilmisany AM, Al-Sawaf H.

http://www.ncbi.nlm.nih.gov/pubmed/16248841

Abstract

Defective sperm function is the most common cause of infertility, and until recently, was difficult to evaluate and treat. Mammalian spermatozoa membranes are rich in **poly unsaturated fatty acids** and are sensitive to oxygen induced damage mediated by lipid peroxidation. Hence, free radicals and reactive oxygen species [ROS] are associated with oxidative stress and are likely to play a number of significant and diverse roles in reproduction. The excessive generation of reactive oxygen species by abnormal spermatozoa and by contaminating leukocytes [leukocytospermia] has been identified as one of the few defined etiologies for male infertility.

Moreover, environmental factors, such as **pesticides, exogenous estrogens, and heavy metals** may negatively impact spermatogenesis since male sperm counts were declined. In addition, aging is also likely to further induce oxidative stress. Limited endogenous mechanisms exist to reverse these damages. In a normal situation, the seminal plasma contains antioxidant mechanisms, which are likely to quench these ROS and protect against any likely damage to spermatozoa. However, during genitourinary infection/inflammation these antioxidant mechanisms may downplay and create a situation called oxidative stress.

Assessment of such oxidative stress status [OSS] may help in the medical treatment of male infertility by suitable antioxidants. The cellular damage in the semen is a result of an improper balance between ROS generation and scavenging activities. Therefore, numerous antioxidants such as **vitamin C, vitamin E, glutathione, and coenzyme Q10**, have proven beneficial effects in treating male infertility. A multi-faceted therapeutic approach to improve male fertility involves identifying harmful environmental and occupational risk factors, while correcting underlying nutritional imbalances to encourage optimal sperm production and function.

The role of antioxidant therapy in the treatment of male infertility

ASHOK AGARWAL & LUCKY H. SEKHON

https://www.clevelandclinic.org/reproductiveresearchcenter/docs/agradoc384.pdf

Abstract
Oxidative stress contributes to defective spermatogenesis leading to male factor infertility. The aim of this study was to review the current literature on the effects of various antioxidants to improve fertilisation and pregnancy rates. The sources of literature were Pubmed and the Cochrane data base. Reviewing the current literature revealed that **Carnitines and vitamin C and E** have been clearly shown to be effective by many well-conducted studies and may be considered as a first line treatment.
The efficacy of antioxidants, such as **glutathione, selenium and coenzyme Q10** has been demonstrated by few, but well-performed studies, and may be considered second line treatment. There is, however, a need for further investigation with randomised controlled studies to confirm the efficacy and safety of antioxidant supplementation in the medical treatment of idiopathic male infertility as well as the need to determine the ideal dose of each compound to improve semen parameters, fertilisation rates and pregnancy outcomes.

http://www.pps.org.pk/PJP/6-2/Raghuveer.pdf

Oxidative stress and role of antioxidants in male infertility

Raghuveer Choudhary, Chawala, VK., Soni ND, Jayant Kumar, Vyas RK.

CONCLUSION
It is crucial for andrologist to understand free radicals, their sources, mechanism of generation, and the damage they can cause to the male reproductive system. In addition, it is also essential to be aware of the various diseases that increase ROS generation in the blood, plasma and seminal fluids.

A multifaceted approach is required for the treatment of male infertility induced by free radicals. Methods can be used to decrease ROS production. (e.g. the addition of antioxidants during Sperm preparation techniques). With the abundance of many synthetic and natural antioxidants, it is very important to use them judiciously. Clinical trials using antioxidants in vivo and in vitro have resulted in a major debate, and further research is required before one can be optimistic about a role for antioxidants in the treatment of infertile man.

Role of male factor in early recurrent embryo loss: do antioxidants have any effect?

http://www.sciencedirect.com/science/article/pii/S0015028208014908#
Aura María Gil-Villa, Wálter Cardona-Maya, Ashok Agarwal, Rakesh Sharma, Ángela Cadavid,

DISCUSSION

The results of this study appear to show that the increased intake of antioxidant-rich foods or antioxidant supplements by men who presented with high levels of DNA damage or oxidative stress improved the gestational results in a group of couples with history of recurrent embryo loss. All couples whose male partners adopted an antioxidant supplementation achieved a successful pregnancy.

Efficacy of Selenium and/or N-Acetyl-Cysteine for Improving Semen Parameters in Infertile Men: A Double-Blind, Placebo Controlled, Randomized Study **Mohammad Reza Safarinejad, Shiva Safarinejad**

http://www.sciencedirect.com/science/article/pii/S0022534708027018

Materials and Methods

The study included 468 infertile men with idiopathic oligo-asthenoteratospermia who were randomized to receive 200 μg selenium orally daily (selenium group of 116), 600 mg N-acetyl-cysteine orally daily (N-acetyl-cysteine group of 118), 200 μg selenium plus 600 mg N-acetyl-cysteine orally daily (selenium plus N-acetyl-cysteine group of 116) or similar regimen of placebo (control group of 118) for 26 weeks, followed by a 30-week treatment-free period. These patients provided blood samples for the measurement of serum testosterone, estradiol, follicle-stimulating hormone, luteinizing hormone, prolactin, inhibin B, selenium and N-acetyl-cysteine. Semen samples were also obtained for routine semen analysis, and the measurement of seminal plasma selenium and N-acetyl-cysteine.

Conclusions

These results indicate that supplemental selenium and N-acetyl-cysteine improve semen quality. We advocate their use for male infertility treatment.

Selenium, a Key Element in Spermatogenesis and Male Fertility

Carla Boitani, Rossella Puglisi

http://link.springer.com/chapter/10.1007/978-0-387-09597-4_4#page-1

ABSTRACT
Selenium is essential for normal spermatogenesis of mammals and its critical role is mainly mediated by two selenoproteins, namely phospholipid hydroperoxide glutathione peroxidase (PHGPx/GPx4) and Selenoprotein P. PHGPx/GPx4 is the major selenoprotein expressed by germ cells in the testis, having multiple functions and representing the pivotal link between selenium, sperm quality and male fertility. Selenoprotein P is a plasma protein that is required for selenium supply to the testis. In the last years, nutritional studies and experimental animal models lacking/ overexpressing a specific PHGPx isoform and selenoprotein P have highly expanded our understanding on how the male reproductive system depends on selenium.

The focus of this review is to report and discuss the most relevant and recent findings in this field. Clinical data have pointed to a correlation between abnormal PHGPx content in sperm and disturbance of human male fertility. However, additional evidence is still required to draw any definitive conclusions about therapeutical strategies for improving fertility by selenium administration.

Searching under 'Glutathione and improving male fertility'

(Glutathione is made through having sufficient ingredients in the body – magnesium, sulphates and Vit C are a start. It cannot be taken orally as the molecules is enormous and is broken down – however can be taken as a Glutathione accelerator).

http://europepmc.org/abstract/MED/10696117/reload=0;jsessionid=R28tSvZa3mVQIpBpsejL.10

Abstract - Studies confirm that male sperm counts are declining, and environmental factors, such as pesticides, exogenous estrogens, and heavy metals may negatively impact spermatogenesis. A number of nutritional therapies have been shown to improve sperm counts and sperm motility, including carnitine, arginine, zinc, selenium, and vitamin B-12. Numerous antioxidants have also proven beneficial in treating male infertility, such as vitamin C, vitamin E, glutathione, and coenzyme Q10. Acupuncture, as well as specific botanical medicines, have been documented in several studies as having a positive effect on sperm parameters. A multi-faceted therapeutic approach to improving male fertility involves identifying harmful environmental and occupational risk factors, while correcting underlying nutritional imbalances to encourage optimal sperm production and function.

The Human Sperm Glutathione System: A Key Role in Male Fertility and Successful Cryopreservation

Meseguer, Marcos; Antonio Martinez-Conejero, Jose; Muriel, Lourdes; Pellicer, Antonio, Remohi, Jose; Garrido, Nicolas

http://www.ingentaconnect.com/content/ben/dml/2007/00000001/00000002/art00006

Abstract:

The equilibrium of the creation and scavenging of free radicals is mandatory in the spermatozoa to fertilize and initiate a full-term pregnancy. The glutathione (GSH) enzymatic system studies have discovered its relationship with oxidative stress in the ejaculate and new strategies to regulate its activity in the semen could be developed. Intracellular sperm GSH system components are altered in infertile men, and these alterations seem to be linked to sperm morphology. We have been able to correlate embryo morphology on 8 cell embryos with the sperm expression of GPx family members; this relationship appears quite promising for discovery of molecular causes of male infertility.

Oxidative stress imbalance potentially leads to damage of the structure of plasma membrane. The freezing and subsequent thawing of sperm is a physically stressful process carried out during routine procedures in assisted reproduction, which results in a highly variable and unpredictable reduction of motile sperm. Subsequently, oxidative status can positively or negatively affect the motility, viability, and fertilizing capacity of thawed sperm. A reserve of glutathione, together with GPx expression, is necessary to eliminate free radicals using GSH or GPx-4 like structural protein and seems to be essential for a good post thaw recovery.

What this does not say is how to get Glutathione. It is made in the body continually and is needed for life. It is the major anti oxidant in the cells and is made from magnesium and sulphur. One quick way to make more and be a lot more vibrant is to in get a glutathione accelerator. Here is the information on how to do this. Perhaps listen to Dr Robert Keller – who discovered how it is possible to take a simple supplement and instantly feel amazing - and especially how to remove modern toxins safely out of the body.

Mammalian glutathione peroxidases control acquisition and maintenance of spermatozoa integrity

E. Chabory, C. Damon,A. Lenoir, J. Henry-Berger, P. Vernet, R. Cadet, F. Saez and J. R. Drevet[2]

http://www.journalofanimalscience.org/content/88/4/1321.short

ABSTRACT

In mammals, post testicular epididymal sperm maturation is considered an essential step in the transformation of immature testicular gametes to mature spermatozoa capable of fertilization. Reactive oxygen species (ROS) have been shown to be key actors in this maturation process, and it is now clear that ROS are central for sperm physiology in processes such as sperm maturation and capacitation. However,

during epididymal maturation and storage and until the onset of fertilization, oxidative damage is a threat spermatozoa must face more than any other cells.

Spermatozoa were found to be extremely sensitive to oxidative attacks correlated with lipid peroxidation, DNA damage, and impaired sperm motility, all affecting fertilization. To control the quantity of H_2O_2 in the vicinity of male gametes, mammalian epididymis uses a panel of nonenzymatic and enzymatic scavengers, among which the **glutathione peroxidase** (GPx) family is largely represented.

Among the various GPx proteins expressed in the mammalian epididymis, GPx4 and GPx5 occupy unique positions and functions that are reviewed in this paper. This paper underlines the importance of the GPx protein family in determining the fertilizing potential of mammalian spermatozoa. This is particularly relevant in the field of mammalian fertility and infertility as well as in the development of assisted medical procreation technologies and male gamete preservation techniques that are extensively used in human and animal reproduction programs.

Treatment of male infertility

Aldo Isidori Maurizio Latini, Francesco Romanelli
http://www.sciencedirect.com/science/article/pii/S0010782405001617#

ABSTRACT

Male factor infertility is a general term that describes a situation in which the inability to conceive is associated with an alteration identified in the male partner. This dysfunction may be associated with low sperm concentration (oligozoospermia), poor sperm motility (asthenozoospermia) or abnormal sperm morphology (teratozoospermia); however, generally, a disturbance of all these variables, oligoasthenoteratozoospermia, is mostly frequent in male subfertility.

For many andrological disorders, it is not possible to find a reasonable cause and various uncontrolled treatments have been applied to infertile men, often just on an empirical basis.

More recently, after the explosive development of modern assisted reproduction techniques (ARTs), feasible with a single spermatozoon [intracytoplasmic sperm injection (ICSI)], **the treatment of male infertility has received new meaning and andrologists are no longer expected to achieve a quantitative increase in sperm number but are instead asked to *improve the fertility potential of the single sperm cell* in order to achieve better results in both in vitro fertilization and ICSI.** Additional prospective studies are needed to better understand the possible role of therapy in ART candidate patients.

Dietary oestrogens and male fertility potential

(Concern over phytyo-estrogens (eg soy) on irreparable times in life)

Mhairi C. L. West[1†], Lorraine Anderson[1], Neil Mcclure[1] and Sheena E. M. Lewis[1]

http://informahealthcare.com/doi/abs/10.1080/14647270500030266

Reports of increased incidences of male reproductive abnormalities and falling sperm counts have prompted interest into the nature of these threats to global fertility. Xenoestrogens have been flagged as major culprits but to date, little is known about the effects of dietary phytoestrogens on male reproductive health. These non-steroidal oestrogens of plant origin are potent endocrine disruptors that modulate normal physiological functions.

Phytoestrogens have become a major component in the typical Western fast food diet over the last few decades. Soy formula milk is another common source of phytoestrogens, now used increasingly as an alternative to breast or cow's milk for infants with allergies. *This use is of particular concern since the most vulnerable periods for oestrogenic insult are thought to be the pre- and neonatal periods when irreversible damage can be inflicted on the developing germinal epithelium*. Studies into the safety of phytoestrogens are urgently needed either to allay fears or increase awareness of the effects of our modern diet on future fertility.

Antioxidant effect of vitamin E on motility, viability and lipid peroxidation of cattle spermatozoa under oxidative stress

Amrit Kaur Bansal, Gurmail Singh
http://www.ighz.edu.pl/files/objects/2647/66/strona5-14.pdf (VIT E)

Vitamin E is one of the major membrane protectants against reactive oxygen species (ROS) and lipid peroxidation (LPO). The study aimed at determining the optimum dose of vitamin E to reverse free radical-mediated oxidative damage on motility, viability and LPO of bulls' sperm. Fresh semen of five local crossbred bulls was suspended in 2.9% sodium citrate, divided into equal fractions and subjected to vitamin E treatment (0, 1, 2, 2.5 mM) in the presence or absence of oxidative stress inducer, i.eferrous ascorbate (FeAA, containing 150 µM FeSOand 750 µM ascorbic acid).

All sperm suspensions were incubated at 37°C for 2 h. Treatment with FeAA reduced sperm motility and viability, but increased the LPO. All doses of vitamin E increased sperm motility and viability, but reduced LPO. However, 2 mM vitamin E was most effective. In conclusion, **vitamin E** reduced the LPO caused by FeAA, and **improved sperm motility and viability** in vitro under induced oxidative stress.

There is plenty of evidence that modern life and choices is impacting in sperm quality - If you really want to know about oxidative stress and fertility – here is a reprint of

http://humupd.oxfordjournals.org/content/14/3/243.full

Oxidative stress and male infertility—a clinical perspective

K Tremellen - 2008

ABSTRACT

Oxidative stress occurs when the production of potentially destructive reactive oxygen species (ROS) exceeds the bodies own natural antioxidant defenses, resulting in cellular damage. Oxidative stress is a common pathology seen in approximately half of all infertile men. ROS, defined as including oxygen ions, free radicals and peroxides are generated by sperm and seminal leukocytes within semen

and produce infertility by two key mechanisms. First, they damage the sperm membrane, decreasing sperm motility and its ability to fuse with the oocyte. Second, ROS can alter the sperm DNA, resulting in the passage of defective paternal DNA on to the conceptus. This review will provide an overview of oxidative biochemistry related to sperm health and will identify which men are most at risk of oxidative infertility. Finally, the review will outline methods available for diagnosing oxidative stress and the various treatments available.

INTRODUCTION

Male factor infertility accounts for up to half of all cases of infertility and affects one man in 20 in the general population (McLachlan and de Kretser, 2001). Evidence now suggests that reactive oxygen species (ROS)-mediated damage to sperm is a significant contributing pathology in 30–80% of cases (Iwasaki and Gagnon, 1992; Zini *et al*., 1993; Ochsendorf, 1994; Shekarriz *et al*., 1995a, b; Agarwal *et al*., 2006a). ROS, defined as including oxygen ions, free radicals and peroxides, cause infertility by two principal mechanisms. First, ROS damage the sperm membrane which in turn reduces the sperm's motility and ability to fuse with the oocyte. Secondly, ROS directly damage sperm DNA, compromising the paternal genomic contribution to the embryo. Despite the common association between compromised sperm quality and oxidative damage, men are rarely screened for oxidative stress nor treated for this condition. Instead they are usually offered 'mechanical' treatments such as intracytoplasmic sperm injection (IVF-ICSI) or intrauterine insemination (IUI). This is less than optimal as oxidative damage to sperm DNA is not directly ameliorated by either IVF-ICSI or IUI treatment. In addition, direct treatment of oxidative stress may allow for natural conception, thereby conserving scarce medical resources. This review will provide an overview of who is at risk of oxidative stress, the mechanisms by which oxidative stress produces infertility and the methods available for its diagnosis and treatment.

OVERVIEW OF OXIDATIVE STRESS BIOCHEMISTRY

ROS are products of normal cellular metabolism. Most of the body's energy is produced by the enzymatically controlled reaction of oxygen with hydrogen in oxidative phosphorylation occurring within the mitochondria during oxidative metabolism. During this enzymatic reduction of oxygen to produce energy, free radicals are formed (Valko *et al*., 2007). A free radical is defined as an oxygen molecule containing one or more unpaired electrons in atomic or molecular orbitals. The addition of one electron to dioxygen (O_2) forms the superoxide anion radical ($O_2^{\bullet -}$), the primary form of ROS. This superoxide anion can then be directly or indirectly (enzymatic, metal catalyzed) converted to secondary ROS such as the hydroxyl radical ($^{\bullet}OH$), peroxyl radical ($ROO^{\bullet}$) or hydrogen peroxide (H_2O_2). The terms free radical and ROS are commonly used in an interchangeable manner, despite the fact that not all ROS are free radicals (Cheeseman and Slater, 1993). For example, hydrogen peroxide (H_2O_2) is considered a ROS but it is not a free radical since it does not contain unpaired electrons. In addition, there is a sub-class of free radicals derived from nitrogen which includes nitrous oxide, peroxynitrite, nitroxyl anion and peroxynitrous acid. Free radicals seek to participate in chemical reactions that relieve them of their unpaired electron, resulting in the oxidation of lipids in membranes, amino acids in proteins and carbohydrates within nucleic acids (Ochsendorf, 1999).

Within semen there are two principal sources of production of free radicals; leukocytes and sperm. The vast majority of semen specimens contain leukocytes,

with neutrophils being the predominant leukocyte type (Aitken *et al*., 1994; Aitken and Baker, 1995). As the production of ROS is one of the principal mechanisms by which neutrophils destroy pathogens, it is not surprising that seminal leukocytes have the potential to cause oxidative stress. However, a link between the presence of leukocytes in semen and male oxidative infertility is still under debate (Wolff, 1995). Several researchers have reported a positive correlation between seminal leukocyte numbers and ROS production (Aitken *et al*., 1994; Whittington *et al*., 1999; Sharma *et al*., 2001). However, other studies have failed to find a significant difference in seminal leukocyte concentration between fertile and infertile men (Christiansen *et al*., 1991; Tomlinson *et al*., 1993; Aitken and Baker, 1995; Rodin *et al*., 2003), and the activation state of leukocytes must also play an important role in determining final ROS output. This is supported by the observation of a positive correlation between seminal ROS production and pro-inflammatory seminal plasma cytokines such as interleukin IL-6 (Camejo *et al*., 2001; Nandipati *et al*., 2005), IL-8 (Rajasekaran *et al*., 1995; Martinez *et al*., 2007) and tumour necrosis factor TNFα (Sanocka *et al*., 2003; Martinez *et al*., 2007).

Every human ejaculate contains leukocytes, which make the quantification of spermatozoa-specific ROS production more complex. However, sperm isolation techniques have been used to confirm that spermatozoa themselves are responsible for some ROS generation, not just contaminating seminal leukocytes (Baker *et al*., 2003). Separation of sperm from seminal leukocytes using density-gradient centrifugation has shown the 'sperm fraction' to produce significant ROS. As this fraction may still contain a very low number of leukocytes, experiments have been conducted where leukocytes are further depleted using magnetic beads coated with leukocyte-specific CD45 antibodies (Aitken *et al*., 1996). After removing all detectable leukocyte contamination, ROS production can still be recorded, confirming the ability of sperm to generate ROS. The relative importance of sperm and leukocyte production of ROS varies between individuals but can be estimated using the leukocyte specific activator, *N*-formyl-methionine-leucine-phenylalanine (FMLP).

The ability of sperm to produce ROS inversely correlates with their maturational state. During spermatogenesis there is a loss of cytoplasm to allow the sperm to form its condensed, elongated form. Immature teratozoospermic sperm are often characterized by the presence of excess cytoplasmic residues in the mid-piece. These residues are rich in the enzyme glucose-6-phosphate dehydrogenase, an enzyme which controls the rate of glucose flux and intracellular production of β-nicotinamide adenine dinucleotide phosphate (NADPH) through the hexose monophosphate shunt. NADPH is used to fuel the generation of ROS via NADPH oxidase located within the sperm membrane (Gomez *et al*., 1996; Fisher and Aitken, 1997; Said *et al*., 2005). As a result, teratozoospermic sperm produce increased amounts of ROS compared with morphologically normal sperm.

The existence of NADPH oxidase activity within sperm was questioned when addition of NADPH was unable to elicit any production of the superoxide anion measured by electron paramagnetic resonance spectroscopy, a very sensitive and specific assay for the superoxide anion (Richer and Ford, 2001). However, since then the presence of a calcium-dependant NADPH oxidase called NOX 5 has been confirmed within sperm (Banfi *et al*., 2001; Armstrong *et al*., 2002; Sabeur and Ball, 2007). This sperm-specific NADPH oxidase (NOX 5) is reported to be quite distinct from leukocyte NADPH oxidase, with NOX 5 activity not being controlled by protein kinase C as occurs in the leukocyte (Armstrong *et al*., 2002). Whether NOX 5 is over expressed in spermatozoa of patients exhibiting infertility associated with oxidative stress is presently unknown.

The relative importance of leukocytes and sperm in the aetiology of oxidative stress is currently under debate. The rate of production of ROS by leukocytes is reported to be 1000 times higher than that of spermatozoa at capacitation (Plante *et al*., 1994), making leukocytes the likely dominant producer of seminal ROS. When seminal ROS production is divided into that produced by the sperm themselves (intrinsic ROS) and that made by the leukocytes (extrinsic), an interesting observation is seen (Henkel *et al*., 2005). While both intrinsic and extrinsic ROS production is negatively correlated with sperm DNA integrity, the relationship is significantly stronger for intrinsic ROS production. This suggests that while leukocytes produce more ROS than sperm on a per cell basis, the close proximity between intrinsic ROS production and sperm DNA makes intrinsic ROS production a more important variable in terms of fertility potential.

The human body has developed several antioxidant strategies to protect itself from ROS damage. This allows for normal oxidative metabolism to occur without damaging the cells, while still allowing for normal ROS-mediated cellular responses such as destruction of infectious pathogens and intracellular signalling (Valko *et al*., 2007). Oxidative stress occurs when the production of ROS overwhelms the antioxidant defense mechanisms leading to cellular damage. Seminal plasma and sperm themselves are well endowed with an array of protective antioxidants (Fujii *et al*., 2003; Garrido *et al*., 2004a). Superoxide dismutase (SOD) and catalase are enzymatic antioxidants which inactivate the superoxide anion ($O^{\bullet}{}_2^-$) and peroxide (H_2O_2) radicals by converting them into water and oxygen. SOD is present within both sperm and seminal plasma (Mennella and Jones, 1980; Zini *et al*., 1993). The addition of SOD to sperm in culture has been confirmed to protect them from oxidative attack (Kobayashi *et al*., 1991). While some investigators have reported minor reductions in seminal plasma SOD activity in infertile men (Alkan *et al*., 1997; Sanocka *et al*., 1997), many have not (Miesel *et al*., 1997; Zini *et al*., 2000; Hsieh *et al*., 2002). However, the majority of evidence does support a link between deficient seminal catalase activity and male infertility (Jeulin *et al*., 1989; Alkan *et al*., 1997; Miesel *et al*., 1997; Sanocka *et al*., 1997; Zini *et al*., 2000). Glutathione peroxidase (GPX) is the final member of the seminal enzymatic antioxidant triad. GPX consists of a family of antioxidants (GPX1-5) that are involved in the reduction of hydroperoxides using glutathione as an electron donor. The GPXs are located within the testis, prostate, seminal vesicles, vas deferens, epididymis, seminal plasma and spermatozoa themselves (reviewed by Vernet *et al*., 2004). GPX must play an important protective role against oxidative attack since its specific inhibition *in vitro* using mercaptosuccinate leads to a large increase in sperm lipid peroxidation (Twigg *et al*., 1998). Male factor infertility has been linked with a reduction in seminal plasma (Giannattasio *et al*., 2002) and spermatozoa (Garrido *et al*., 2004b) GPX activity, further supporting an important role for this enzyme in male fertility. In addition, men exhibiting leukospermia-associated oxidative stress have been reported to have significantly reduced GPX activity within their spermatozoa (Therond *et al*., 1996). Finally, the continued activity of GPX depends on the regeneration of reduced glutathione by glutathione reductase (GTR). Selective inhibition of GTR reduces the availability of reduced glutathione for maintaining GPX activity, thereby exposing sperm to oxidative stress (Williams and Ford, 2004). The coordinated activity of GPX, GTR and glutathione clearly play a pivotal role in protecting sperm from oxidative attack.

The non-enzymatic antioxidants present within semen include ascorbic acid (Vitamin C), α-tocopherol (Vitamin E), glutathione, amino acids (taurine, hypotaurine), albumin, carnitine, carotenoids, flavenoids, urate and prostasomes. These agents principally act by directly neutralizing free radical activity chemically. However, they

also provide protection against free radical attack by two other mechanisms. Albumin can intercept free radicals by becoming oxidized itself, thereby sparing sperm from attack (Twigg *et al*., 1998). Alternatively, extracellular organelles (prostasomes) secreted by the prostate have been shown to fuse with leukocytes within semen and reduce their production of free radicals (Skibinski *et al*., 1992; Saez *et al*., 1998). A substantial number of researchers have reported a significant reduction in non-enzymatic antioxidant activity in seminal plasma of infertile compared with fertile men (Fraga *et al*., 1991; Fraga *et al*., 1996; Smith *et al*., 1996; Therond *et al*., 1996; Lewis *et al*., 1997; Gurbuz *et al*., 2003; Koca *et al*., 2003; Mostafa *et al*., 2006; Song *et al*., 2006).

Antioxidants contained within seminal plasma are obviously helpful for preventing sperm oxidative attack following ejaculation. However, during spermatogenesis and epididymal storage, the sperm are not in contact with seminal plasma antioxidants and must rely on epididymal/testicular antioxidants and their own intrinsic antioxidant capacity for protection. Sperm are therefore vulnerable to oxidative damage during epididymal transit, especially when there is epididymal inflammation such as male genital tract infection. In addition, testicular biopsies from men with varicocele-associated oxidative stress have shown an increase in oxidative DNA damage within spermatogonia and spermatocytes (Ishikawa *et al*., 2007). Therefore, while seminal plasma antioxidants may help minimize ejaculated sperm oxidative stress, they have no capacity to prevent oxidative damage initiated ‘up stream’ at the level of the testis and epididymis.

SEMINAL FREE RADICALS—FRIEND OR FOE?

Sperm were the first type of cell reported to produce free radicals. In this pioneering report, MacLeod (1943) noted that incubation of sperm under conditions of high oxygen tension lead to a rapid loss of their motility. The addition of the antioxidant catalase to the medium preserved sperm motility, prompting MacLeod to suggest that sperm must produce hydrogen peroxide during normal oxidative metabolism. Since this publication, it has evolved that three inter-related mechanisms account for oxidative stress-mediated male infertility—impaired motility, impaired fertilization and oxidative DNA damage.

The underlying pathology behind free radicals ability to reduce sperm motility was first reported by Jones *et al*. (1979). They reported that ROS-induced peroxidation of the sperm membrane decreasing its flexibility and therefore tail motion. Sperm membranes are vulnerable to this type of damage as they contain large amounts of unsaturated fatty acids. Direct ROS damage to mitochondria, decreasing energy availability, may also impede sperm motility (de Lamirande and Gagnon, 1992; de Lamirande *et al*., 1997, 1998). By either mechanism, oxidative stress impairs sperm motility and will result in less sperm reaching the oocyte for fertilization (Whittington *et al*., 1999; Kao *et al*., 2007).

Low level production of free radicals by sperm plays a positive role in preparation for fertilization (capacitation). Hydrogen peroxide stimulates the acrosome reaction and sperm hyperactivation (de Lamirande and Gagnon, 1993), thereby assisting the sperm's transit through the cumulus and zona pellucida. Low concentrations of hydrogen peroxide also cause tyrosine phosphorylation, which augments sperm membrane binding to the zona pellucida ZP-3 protein (Aitken *et al*., 1995b), ultimately boosting sperm–oocyte fusion (Aitken *et al*., 1998). However, high levels of ROS production lead to peroxidation of the sperm acrosomal membrane and diminished acrosin activity (Zalata *et al*., 2004), and impaired sperm–oocyte fusion

(Aitken *et al*., 1989; Ichikawa *et al*., 1999; Saleh *et al*., 2003a, b; Zorn *et al*., 2003a; Jedrzejczak *et al*., 2005).

Free radicals have the ability to directly damage sperm DNA by attacking the purine and pyrimidine bases and the deoxyribose backbone. Normally, sperm DNA is tightly packaged by protamines protecting it from free radical attack. However, infertile men often exhibit deficient protamination, leaving the sperm DNA particularly vulnerable to ROS attack (Oliva, 2006). Alternatively, free radicals can initiate apoptosis within the sperm, leading to caspase-mediated enzymatic degradation of the DNA (Kemal Duru *et al*., 2000; Wang *et al*., 2003; Moustafa *et al*., 2004; Villegas *et al*., 2005). Several investigators (Kodama *et al*., 1997; Aitken *et al*., 1998; Saleh *et al*., 2002b; Oger *et al*., 2003; Wang *et al*., 2003; Henkel *et al*., 2005; Kao *et al*., 2007) have now confirmed the link between oxidative stress and sperm DNA damage using various techniques such as terminal deoxynucleotidyl transferase-mediated dUTP nick-end labeling (TUNEL), sperm chromatin structure assay (SCSA) and measurement of the byproduct of DNA oxidation, 8-hydroxydeoxyguanosine (8-OHdG). Furthermore, two groups have now correlated increased sperm oxidative DNA damage with poor blastocyst formation *in vitro* (Zorn *et al*., 2003a; Meseguer *et al*., 2006, 2007). Damaged paternal DNA is recognized to be a significant cause for poor blastocyst development (Seli *et al*., 2004). Finally, a large prospective study of 225 couples planning their first pregnancy found a strong inverse relationship between seminal 8-OHdG concentration and monthly natural fecundity (Loft *et al*., 2003).

During natural conception or routine IVF, oxidative damage to the sperm membrane will normally block fertilization, preventing the damaged paternal DNA from creating an embryo. However, during IVF-ICSI this natural barrier to fertilization is lost and sperm containing significantly damaged DNA can still achieve fertilization following microinjection (Zorn *et al*., 2003a). While many of these embryos will ultimately fail at the blastocyst or early fetal stage, there is the potential for a child to be born with damaged paternal derived DNA. The consequences of this are as yet unknown but it has been suggested to include the initiation of genetic defects and childhood cancer (Aitken and Krausz, 2001; Aitken *et al*., 2003).

ORIGINS OF OXIDATIVE STRESS

The origins of sperm oxidative stress are summarized in Fig. 1. While pathologies such as genitourinary tract infection and varicocele are well established causes of oxidative stress, others such as hyper-homocysteinaemia and diabetes are only now just becoming recognized as possible causes. It is hoped that this review will stimulate further research in these less well established potential causes of male oxidative infertility.

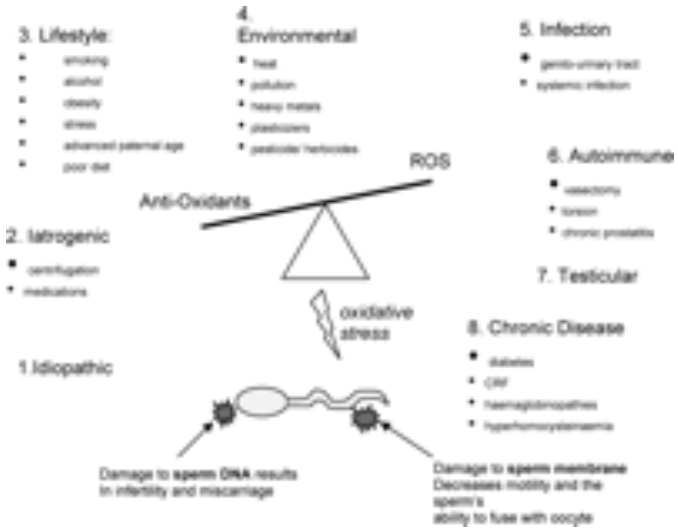

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Figure 1:

The oxidative stress balance.

Idiopathic

Idiopathic male factor infertility has been linked with oxidative stress by several research groups. One of the principal causes of this association is the observation that morphologically abnormal sperm have an increased capacity to generate ROS, but also a reduced antioxidant capacity (Gomez *et al.*, 1996; Garrido *et al.*, 2004b; Said *et al.*, 2004; Said *et al.*, 2005). As approximately one-third of infertile men exhibit teratozoospermia (Thonneau *et al.*, 1991), it is not surprising that sperm oxidative stress is commonly identified in the idiopathic infertile male population. Even men with normozoospermic idiopathic infertility exhibit significantly higher seminal ROS production and lower antioxidant capacity than fertile men (Pasqualotto *et al.*, 2001; Agarwal *et al.*, 2006b), for as yet unknown reasons.

Iatrogenic

The use of assisted reproductive technologies (ART) has the potential to exacerbate sperm oxidative stress. During IVF and IUI treatment semen is centrifuged to separate sperm from seminal plasma. This exacerbates oxidative stress as centrifugation increases sperm ROS production many fold (Iwasaki and Gagnon, 1992; Shekarriz *et al.*, 1995a, b), while removing sperm from protective antioxidants within seminal plasma (Potts *et al.*, 2000a, b). In addition cryopreservation of sperm, another commonly used technique in ART, is associated with an increase in sperm oxidative stress (Watson, 2000).

Drugs such as the chemotherapy agent cyclophosphamide have been linked with sperm oxidative stress. Administration of cyclophosphamide to animals is reported to increase testicular malondialdehyde (MDA) levels and produce a fall in testicular catalase, implying the presence of oxidative stress (Das *et al.*, 2002; Ghosh *et al.*, 2002). Drugs such as aspirin and paracetamol (acetaminophen) can also produce oxidative stress by increasing cytochrome P450 activity, thereby boosting ROS generation (Agarwal and Said, 2005).

Lifestyle

Smoking results in a 48% increase in seminal leukocyte concentrations and a 107% increase in seminal ROS levels (Saleh *et al*., 2002a). Smokers have decreased levels of seminal plasma antioxidants such as Vitamin E (Fraga *et al*., 1996) and Vitamin C (Mostafa *et al*., 2006), placing their sperm at additional risk of oxidative damage. This has been confirmed by the finding of a significant increase in levels of 8-OHdG within smoker's seminal plasma (Fraga *et al*., 1996).

Dietary deficiencies have been linked with sperm oxidative damage by several research groups. The Age and Genetic Effects in Sperm (AGES) study examined the self-reported dietary intake of various antioxidants and nutrients (vitamins C and E, β-carotene, folate and zinc) in a group of 97 healthy non-smokers and correlated this with sperm quality (Eskenazi *et al*., 2005). This study did observe a significant **correlation between vitamin C intake and sperm concentration and between vitamin E intake and total progressively motile sperm.** This is also consistent with earlier reports of a significant link between seminal plasma vitamin E levels and an increase in percentage of motile sperm (Therond *et al*., 1996).

However, the AGES study was unable to confirm a link between low intake of antioxidants and sperm DNA damage (Silver *et al*., 2005). This was surprising given that other researchers had linked low seminal plasma vitamin C levels with increased sperm DNA damage (Fraga *et al*., 1991; Song *et al*., 2006). It is possible that levels of individual antioxidants within seminal fluids may more accurately reflect biological effect than self-reported dietary intake as different food sources and preparation techniques can vastly modify antioxidant intake.

Alternatively, differences in the populations studied may explain the discrepant results. Song *et al*. (2006) correlated sperm DNA damage with dietary antioxidant intake in infertile men, while Silver *et al*. (2005) and Fraga *et al*. (1991) examined this relationship in healthy presumed fertile patients. Fertile men with low levels of oxidative attack may not be as dependant on seminal antioxidants for protection of their sperm DNA integrity. Therefore, a dietary deficiency in antioxidants may not lead to sperm oxidative DNA damage in this fertile cohort.

Excessive alcohol consumption causes an increase in systemic oxidative stress as ethanol stimulates the production of ROS, while many alcohol abusers have diets deficient in protective antioxidants (Wu and Cederbaum, 2003; Koch *et al*., 2004). A study of 46 alcoholic men of reproductive age has suggested the presence of oxidative stress within the testicle by reporting a significant reduction in plasma testosterone, increase in serum lipid peroxidation byproducts and a drop in antioxidants (Maneesh *et al*., 2006). However, no study to date has directly examined the link between alcohol intake and sperm oxidative damage.

Extremes of exercise activity, at both ends of the spectrum, have been linked with oxidative stress. It is not surprising that high impact exercise is linked with oxidative stress since muscle aerobic metabolism creates a large amount of ROS (Peake *et al*., 2007). In a rodent model, increasing levels of exercise are linked with a reduction in sperm count and motility and a corresponding increase in biochemical signs of testicular oxidative stress (Manna *et al*., 2004). Conversely, obesity produces oxidative stress as adipose tissue releases pro-inflammatory cytokines that increase leukocyte production of ROS (Singer and Granger, 2007). Furthermore, accumulation of adipose tissue within the groin region results in heating of the

testicle which has been linked with oxidative stress and reduced sperm quality (Banks *et al*., 2005; Ishii *et al*., 2005; Perez-Crespo *et al*., 2007).

Psychological stress produces a reduction in semen quality; with the underlying mechanism previously felt to be related to a central impairment of gonadotrophin drive (Fenster *et al*., 1997). However, recent prospective studies have linked a period of psychological stress with a reduction in sperm quality mediated by an increase in seminal plasma ROS generation and a reduction in antioxidant protection (Eskiocak *et al*., 2005, 2006).

Several studies have reported that **sperm DNA damage increases with advancing age in both fertile** (Wyrobek *et al*., 2006) and infertile men (Singh *et al*., 2003; Moskovtsev *et al*., 2006). It is possible that an increase in oxidative sperm DNA damage is the underlying pathology. A large observational study has confirmed that systemic oxidative stress increases with age (Junqueira *et al*., 2004). Animal studies using the Brown Norway rat, an established model of male reproductive aging, confirm that sperm from older animals produce more free radicals than from young animals and have a reduced enzymatic antioxidant activity, resulting in an increase in ROS-mediated sperm DNA damage (Zubkova *et al*., 2005; Weir and Robaire, 2007).

Environmental

Phthalates are chemicals used as a plastics softener and are contained in a wide range of food packaging and personal care products. Exposure to phthalates can occur via dietary consumption, dermal absorption or inhalation and has been linked with impaired spermatogenesis and increased sperm DNA damage (Agarwal *et al*., 1985; Srivastava *et al*., 1990; Kasahara *et al*., 2002; Hauser *et al*., 2007). Oral administration of phalate esters to rats is reported to increase the generation of ROS within the testis and a concomitant decrease in antioxidant levels, culminating in impaired spermatogenesis (Lee *et al*., 2007).

Several environmental pollutants have been linked with testicular oxidative stress. Pesticides such as lindane (Chitra *et al*., 2001), methoxychlor (Latchoumycandane *et al*., 2002) and the herbicide dioxin-TCDD (Latchoumycandane *et al*., 2003) have all been linked with testicular oxidative stress in rodent models. The commonly used preservative sulfur dioxide has also been shown to produce testicular oxidative stress in laboratory animals (Meng and Bai, 2004). Air pollutants such as diesel particulate matter act as potent stimuli for leukocyte ROS generation (Gonzalez-Flecha, 2004; Alaghmand and Blough, 2007). While no study has directly linked airborne pollutants with testicular oxidative stress, it is possible that this oxidative insult is responsible for the increase in sperm DNA damage seen following periods of airborne pollution (Rubes *et al*., 2005).

Heavy metal exposure has been conclusively linked with sperm oxidative damage. Both cadmium and lead are linked with an increase in testicular oxidative stress (Hsu and Guo, 2002; Acharya *et al*., 2003) and a resultant increase in sperm DNA oxidation (Xu *et al*., 2003; Naha and Chowdhury, 2006). The increase in infertility and miscarriage observed in the partners of welders and battery/paint factory workers (Gennart *et al*., 1992; Bonde, 1993) may be due to oxidative damage to sperm DNA initiated by the inhalation of metal fumes.

Infection

Genitourinary tract infection

Up to 50% of men will experience prostatitis at some point in their lives, with prostatitis becoming chronic in 10% of men (Schaeffer, 2003). Bacteria responsible for prostate infection may originate from the urinary tract or can be sexually transmitted (Fraczek and Kurpisz, 2007; Fraczek *et al*., 2008). Typical non-sexually-transmitted pathogens include *Streptococci* (*S. viridans* and *S. pyogens*), coagulase-negative *Staphylococci* (*S. epidermidis*, *S. haemolyticus*), gram-negative bacteria (*E. coli*, *Proteus mirabilis*) and atypical mycoplasma strains (*Ureaplasma urealyticum*, *Mycoplasma hominis*). All of these pathogens will create an acute inflammatory response with an influx of leukocytes into the genital tract and a resulting increase in ROS production (Mazzilli *et al*., 1994; Depuydt *et al*., 1996; Ochsendorf, 1999; Potts *et al*., 2000a, b). Men prone to recurrent genitourinary tract infections, such as paraplegics, have been confirmed to have high degrees of sperm oxidative pathology (Padron *et al*., 1997; Brackett *et al*., 2008). Current or past *Chlamydia* infection has also been linked with an increase in oxidative damage to sperm (Segnini *et al*., 2003).

Viral infections may also initiate oxidative damage to sperm. The link between common viral pathogens such as cytomegalovirus, herpes simplex virus (HSV), Epstein-Barr virus and oxidative infertility has been examined by several groups. Only HSV appears to have a possible role in the initiation of oxidative damage to sperm. Herpes simplex DNA is found in 4–50% of infertile men's semen (Kapranos *et al*., 2003, Bezold *et al*., 2007), with IgM antibodies towards HSV being associated with a 10-fold increase in the rate of leukospermia (Krause *et al*., 2002, 2003). Given the well recognized link between leukospermia and seminal ROS levels, together with the observation of a reduction in sperm motility in men positive for seminal HSV DNA (Kapranos *et al*., 2003), it is likely that HSV is a viral pathogen involved in oxidative stress.

Systemic infection

Several chronic systemic infections have been linked with increased oxidative stress throughout the body. Human immunodeficiency virus (HIV) infection is associated with an increase in leukocyte number and activation within semen (Umapathy *et al*., 2001). Hepatitis B and C infection has also been correlated with significant hepatic oxidative stress (Chen and Siddiqui, 2007; Seronello *et al*., 2007). At present it is unknown if this oxidative stress extends to the semen, but impaired sperm motility seen in hepatitis B and C patients (Durazzo *et al*., 2006; Vicari *et al*., 2006), makes this possible. Finally, chronic infections such as tuberculosis (Srinivasan *et al*., 2004), leprosy (Vijayaraghavan *et al*., 2005), malaria (Guha *et al*., 2006) and Chagas disease (Macao *et al*., 2007) have all been linked with elevated degrees of systemic oxidative stress. While no study has directly linked these chronic infectious diseases with sperm oxidative stress, it is unlikely that the male reproductive tract would be spared from this systemic oxidative insult.

Autoimmune/inflammatory

Chronic non-bacterial prostatitis (NIH Category III) is a chronic inflammation of the prostate in the absence of infection and has been reported by several groups to be associated with considerably elevated oxidative stress within semen (Pasqualotto *et al*., 2000; Shahed and Shoskes, 2000; Potts and Pasqualitis, 2003). Chronic non-

bacterial prostatitis accounts for in excess of 90% of all cases and effects 10% of men (Schaeffer, 2003). In the majority of cases of chronic non-bacterial prostatitis it is reported that an adverse autoimmune response to seminal or prostate antigens is responsible for the pathology, leading to an increase in pro-inflammatory cytokines and activated ROS producing leukocytes within the semen (Batstone *et al*., 2002; Motrich *et al*., 2005; Motrich *et al*., 2007). While the exact trigger for this response is unknown, one report has linked a polymorphism of the TH-2 cytokine IL-10 with chronic non-bacteria prostatitis (Shoskes *et al*., 2002). A lack of this Th-2 cytokine may tip the immune balance towards the Th-1 direction leading to the generation of T lymphocytes reactive against prostate antigens. These T cells will liberate cytokines such as IFN-γ, TNF-α and IL-1β that stimulate chemotaxis and activation of leukocytes, leading to increased seminal oxidative stress (Motrich *et al*., 2005). It is therefore not surprising to see the majority of studies linking chronic non-bacterial prostatitis with a significant reduction in sperm density, motility, morphology and membrane integrity (Christiansen *et al*., 1991; Leib *et al*., 1994; Krieger *et al*., 1996; Engeler *et al*., 2003; Motrich *et al*., 2005; Henkel *et al*., 2006); although this is refuted by some groups (Pasqualotto *et al*., 2000; Ludwig *et al*., 2003).

Oxidative stress has been proposed as a significant cause for infertility after vasectomy reversal. It is believed that vasectomy disrupts the normal blood-testis barrier, leading to a loss of immune privilege and activation of immune responses against sperm (Filippini *et al*., 2001). Several studies have documented an increase in seminal leukocytes, pro-inflammatory cytokines and free radical production within semen following vasectomy reversal (Shapiro *et al*., 1998; Kolettis *et al*., 1999; Sharma *et al*., 1999; Nandipati *et al*., 2005).

Testicular

Oxidative stress is now widely believed to be the principal underlying pathology linking **varicocele with male infertility** (Hendin *et al*., 1999; Barbieri *et al*., 1999; Saleh *et al*., 2003b; Nallella *et al*., 2004; Smith *et al*., 2006; Agarwal *et al*., 2006c; Ishikawa *et al*., 2007; Smith *et al*., 2007). The increase in varicocele-related ROS production is strongly correlated with a reduction in sperm DNA integrity when assessed by either TUNEL (Smith *et al*., 2006) or 8-hydroxy-2′-deoxyguanosine DNA oxidative metabolite levels (Chen *et al*., 2004).

Cryptorchidism is a common cause for male factor infertility in which the primary pathology is hypo-spermatogenesis due to deficient maturation of gonocytes to type A spermatogonia (Huff *et al*., 1991). However, recently it has been reported that men with cryptorchidism surgically treated with orchidoplexy early in life still have markedly elevated sperm ROS production and DNA fragmentation compared with fertile controls (Smith *et al*., 2007).

Torsion of the spermatic cord has long been recognized as a cause of male infertility, even when this torsion is unilateral. It is now generally accepted that oxidative stress related to ischemia-reperfusion injury is the underlying cause of damage to both the torted and contra-lateral testis. A prolonged period of ischemia followed by surgical or spontaneous restoration of blood flow leads to an influx of activated leukocytes into both testis (Turner *et al*., 2004) and a consequent increase in generation of free radicals (Filho *et al*., 2004). Oxidative stress then leads to necrosis of the germinal cells with resulting subfertility or infertility.

Chronic disease

Diabetes has long been recognized to impair male fertility by interfering with both spermatogenesis and erectile function. Recently it has been reported that diabetic men have significantly higher levels of sperm DNA fragmentation than normal controls (Agbaje *et al*., 2007). While this study did not directly measure oxidative stress, the authors proposed that the most likely mechanism for the observed increase in sperm DNA damage was an increase in oxidative stress as this is now recognized as a key pathology underlying many chronic complications of diabetes. In support, studies using the Streptozotocin-induced diabetic rat model have found a significant increase in testicular oxidative stress within 6 weeks of initiation of the diabetic state (Shrilatha and Muralidhara, 2007).

Chronic inflammation and oxidative stress are highly prevalent in patients with chronic kidney disease and end-stage renal disease (Oberg *et al*., 2004). Surprisingly, even when uraemia is reversed by haemodialysis, a persisting state of chronic inflammation and oxidative stress persists (Danielski *et al*., 2003; Pupim *et al*., 2004). Furthermore, renal transplant patients with stable renal function and no obvious signs of immune rejection of their graft also have elevated levels of oxidative stress (Moreno *et al*., 2005).

Patients with haemaglobinopathies such as beta-thalassemia major have high degrees of systemic oxidative stress (Livrea *et al*., 1996), with this oxidative damage confirmed to involve sperm (Carpino *et al*., 2004). The likely cause of oxidative stress is iron overload from multiple blood transfusions. Iron is a potent pro-oxidant capable of redox cycling when not safely bound to transferrin in the blood or stored as ferritin in tissue.

The toxic accumulation of homocysteine may cause reproductive dysfunction and oxidative stress within the testis (Forges *et al*., 2007; Sonmez *et al*., 2007). Hyper-homocysteinaemia usually occurs due to suboptimal re-methylation of homocysteine to methionine by the enzyme methyl tetrahydrofolate reductatse (MTHFR) caused by a dietary deficiency of folate or a single-nucleotide polymorphism (SNP) in the MTHFR gene (Selhub, 1999; Matthews, 2002). Several investigators have reported that SNPs (C677T and others) in the MTHFR gene are more commonly found in the infertile men (Bezold *et al*., 2001; Park *et al*., 2005; Lee *et al*., 2006; Zhou-Cun *et al*., 2007), placing these men at increased risk of homocysteine-induced oxidative stress.

LABORATORY IDENTIFICATION OF OXIDATIVE STRESS-RELATED MALE INFERTILITY

One of the main reasons why screening for oxidative stress is not routine in andrology laboratories is the cost and complexity of testing and the lack of a single standardized measure of oxidative stress. At present there are over 30 assays of oxidative stress (Ochsendorf, 1999), broadly divided into three different types. This review will focus on the most popular and clinically useful assays currently being performed.

Direct methods

These assays measure damage created by excess free radicals against the sperm lipid membrane or DNA. As oxidative stress is the result of an in balance between ROS production and total antioxidant capacity (TAC), direct tests reflect the net biological effect between these two opposing forces.

The most widely used method of assessing sperm membrane peroxidation is the measurement of MDA levels in sperm or seminal plasma with the thiobarbituric acid assay. MDA levels in sperm are quite low and therefore require the use of sensitive high-pressure liquid chromatography (HPLC) equipment (Li *et al*., 2004; Shang *et al*., 2004) or the use of iron-based promoters and spectrofluometry measurement (Aitken *et al*., 1993). Seminal plasma levels of MDA are 5–10-fold higher than sperm, making measurement on standard spectrophotometers possible (Sanocka *et al*., 1997; Nakamura *et al*., 2002; Tavilani *et al*., 2005). Measurement of MDA appears to be of some clinical relevance since its concentration within both seminal plasma and sperm is elevated in infertile men with excess ROS production, compared with fertile controls or normozoospermic individuals (Sanocka *et al*., 1997; Nakamura *et al*., 2002; Tavilani *et al*., 2005; Hsieh *et al*., 2006). Furthermore, *in vitro* impairment of motility, sperm DNA integrity and sperm–oocyte fusion capacity by ROS is accompanied by an increase in MDA concentration (Aitken *et al*., 1989, 1993). Other direct tests of sperm membrane lipid peroxidation such as measurement of the isoprostane 8-Iso-PGF2α (Khosrowbeygi and Zarghami, 2007) and the c11-BODIPY assay (Aitken *et al*., 2007; Kao *et al*., 2007) are showing promise but are not yet in common usage.

It is well recognized that oxidative stress is one of the major causes of sperm DNA damage (Aitken *et al*., 1998; Oger *et al*., 2003; Saleh *et al*., 2003a, b). However, measurement of sperm DNA damage by TUNEL or SCSA is an imperfect assessment of oxidative stress as sperm DNA can be damaged by non-oxidative mechanisms such as aberrant apoptosis and incomplete sperm protamination (Ozmen *et al*., 2007). The best direct assessment of sperm DNA oxidative damage is the measurement of the oxidized deoxynucleoside, 8-oxo-7,8-dihydro 2′ deoxyguanosine (8-OHdG). This can be measured in sperm or seminal plasma by HPLC (Fraga *et al*., 1991; Loft *et al*., 2003), enzyme-linked immunoabsorbent assay (Nakamura *et al*., 2002) or directly within sperm using immunoflurorescence (Kao *et al*., 2007). Since a large prospective study has reported that chances of natural conception is inversely correlated with sperm 8-OHdG levels (Loft *et al*., 2003), measurement of this direct marker of sperm oxidative stress appears to have some clinical utility.

Indirect methods

Chemoluminescence assays using either Luminol or Lucigenin are the most commonly described technique to detect ROS production within semen. These probes are very sensitive and have the advantage of relatively well established reported ranges for both the fertile and infertile population (Ochsendorf *et al*., 1994; Williams and Ford, 2005; Athayde *et al*., 2007). However, general uptake by clinical andrology laboratories has been hampered by expensive equipment (luminometer) and difficulties with quality control created by assay confounders such as incubation time, leukocyte contamination and presence of seminal plasma contamination (Kobayashi *et al*., 2001; Aitken *et al*., 2004). Furthermore, Lucigenin has been shown to undergo auto-oxidization which itself leads to the production of superoxide anions (Liochev and Fridovich, 1997). This makes chemoluminescent probes such as Lucigenin less than ideal reagents for measurement of sperm superoxide anion production. A simpler alternative may be light microscopy quantification of nitroblue tetrazolium (NBT) activity. NBT is a yellow water soluble compound that reacts with superoxide anions within cells to produce a blue pigment diformazan. The amount of diformazan crystals seen within a leukocyte or sperm reflects its superoxide anion production. The NBT assay has been shown to correlate well with traditional chemoluminescence techniques (Esfandiari *et al*., 2003) but has two distinct

advantages. First, the NBT assay is inexpensive to set up as it only requires a light microscope. Secondly, the NBT assay can discriminate between production of ROS by sperm and leukocytes without the need for addition of activating peptides (FMLP) used in chemoluminescence assays (WHO manual, 1999).

Measurement of TAC within semen can be conducted in a variety of ways. The ability of seminal plasma to inhibit chemoluminescence elicited by a constant source of ROS (horse-radish peroxidase) is a commonly used technique. The TAC is usually quantified against a Vitamin E analogue (Trolox) and expressed as a ROS-TAC score (Sharma *et al*., 1999). However, colourimetry techniques based on the colour change of ABTS (2,2′-azinobis3-ethylbenzo-thiazoline-6-sulphate) are now becoming more popular as they are cheaper and easier to perform (Said *et al*., 2003; Erel, 2004). The reduced ABTS molecule is oxidized to ABTS+ using hydrogen peroxide and a peroxidase to form a relatively stable blue-green colour measured at 600 nm with a standard spectrophotometer. Antioxidants present within seminal plasma suppress this colour change to a degree that is proportional to their concentrations. Again the antioxidant activity is quantified using Trolox.

Oxidative stress implied from routine semen analysis

A summary of the routine laboratory test 'sentinel signs' suggesting the possible presence of sperm oxidative stress is contained in Table I. While a reduction in any of the sperm parameters (count, motility, morphology) is more commonly seen in men with oxidative stress, asthenozoospermia is probably the best surrogate marker for oxidative stress in a routine semen analysis (Aitken and Baker, 1995; Aitken *et al*., 1995a, b; Whittington *et al*., 1999; Keskes-Ammar *et al*., 2003; Kao *et al*., 2007). A link between impaired sperm motility and oxidative stress also extends to the sperm DNA as a recent study has identified a highly significant correlation between oxidation of sperm DNA and reduced motility (Kao *et al*., 2007).

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Table I.

Sentinel laboratory signs suggesting possible sperm oxidative stress.

Hyperviscosity of seminal plasma is associated with increased levels of seminal plasma MDA (Aydemir *et al*., 2008) and reduced seminal plasma antioxidant status (Siciliano *et al*., 2001), making impaired viscosity a reasonable surrogate marker of oxidative stress. Infection of the semen with *Ureaplasma urealyticum* is associated with increased seminal plasma viscosity (Wang *et al*., 2006) and an increase in ROS production (Potts *et al*., 2000a, b). It is possible that these infections may damage the prostate and seminal vesicle, altering the substrates required for creation of normal semen viscosity.

A large number of round cells within semen may suggest the presence of oxidative stress as they may represent seminal leukocytes (Sharma *et al*., 2001). However, round cells may also be immature sperm rather than leukocytes, so formal identification of leukocytes requires ancillary tests such as the peroxidase test, CD45 staining or measurement of seminal elastase (WHO manual, 1999; Zorn *et al*., 2003b; Kopa *et al*., 2005). Finally, poor sperm membrane integrity assessed by the hypo-osmolar swelling test has been linked with the presence of sperm oxidative stress (Dandekar *et al*., 2002).

MANAGEMENT OF OXIDATIVE STRESS RELATED INFERTILITY

Once an individual has been identified as having oxidative stress related infertility, treatment should be aimed at identification and amelioration of the underlying cause before considering antioxidant treatment. The following paragraphs are the author's suggestions for investigation and management based on the underlying causes of oxidative stress outlined in previous paragraphs. These recommendations are summarized in Table II.

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Table II.

Summary of treatment options in male oxidative infertility.

Lifestyle modification

Lifestyle behaviours such as smoking, poor diet, alcohol abuse, obesity or psychological stress have all been linked with oxidative stress. While the effectiveness of elimination of these lifestyle triggers for oxidative stress has not been formally tested, it is likely that making positive lifestyle changes such as a diet high in fruit/vegetables, maintenance of normal weight and a reduction in smoking /alcohol intake would have at least some beneficial effect on sperm health.

Environmental exposures

Exposure to heat, pollution and toxins (heavy metals and plasticizers) have all been linked with oxidative stress. Men should be advised to avoid activities which may heat the scrotum such as long baths and saunas. Proper ventilation and use of personal protective equipment at work will hopefully reduce men's exposure to chemical and metal vapours linked with oxidative stress.

Treatment of infection/inflammation

Infection of the semen and male accessory sex glands with *Chlamydia* and *Ureaplasma* has been conclusively linked with an increase in oxidative stress. As both of these infections are treatable with antibiotics, it makes sense to screen all men with known oxidative stress for these bacterial pathogens. Two studies have now confirmed the ability of antibiotic treatment to reduce sperm oxidative stress and subsequently improve sperm quality (Omu *et al.*, 1998; Vicari, 2000). One relatively large and well-conducted study randomized men with *Chlamydia* or *Ureaplasma* infection to either 3 months of antibiotics or no treatment (Vicari, 2000). Compared with the controls, the antibiotic treated group exhibited a significant fall in seminal leukocytes and ROS production at 3 months, an improvement in sperm motility and a significant improvement in natural conception (28.2 versus 5.4%, $P = 0.009$). A smaller study using only 10 days of antibiotic treatment did not produce any significant decline in seminal leukocyte count or improvement in motility (Krause *et al.*, 2003). While this study did not measure ROS production in semen, it is likely that prolonged courses of antibiotics (3 months) are required to completely irradiate difficult-to-treat male accessory gland infections and reverse oxidative pathology.

In addition to antibiotic treatment, non-steroidal anti-inflammatory (NSAID) drugs may also reduce seminal leukocytes production of free radicals. In one study men with

antibiotic treated *Chlamydia* or *Ureaplasma* infection were randomized to either a NSAID or carnitine antioxidant and monitored for improvements in sperm quality over the next 4 months (Vicari *et al*., 2002). Those men treated with 2 months of NSAID followed by 2 months of carnitine had the most significant reduction in seminal ROS production and improvement in sperm motility/viability. In addition, a one month course of a COX-2 anti-inflammatory has been shown to significantly reduce sperm leukocyte count, while improving sperm motility, morphology and viability (Gambera *et al*., 2007). It would therefore appear that a combination of antibiotics followed by a course of anti-inflammatory medication is the preferred treatment path in infection related oxidative stress.

Direct treatment of oxidative pathology

Several investigators have reported that surgical treatment of a varicocoele can reduce seminal ROS levels and improve sperm DNA integrity (Mostafa *et al*., 2001; Zini *et al*., 2005; Hurtado de Catalfo *et al*., 2007; Werthman *et al*., 2007). While the most recent meta-analysis examining the effect of varicocelectomy on spontaneous conception shows a significant benefit (Marmar *et al*., 2007), the Cochrane Database suggests that there is no benefit (Evers and Collins, 2004). Well-conducted randomized studies measuring oxidative end-points (sperm lipid peroxidation and oxidative DNA damage) and pregnancy rates need to be performed before routine use of varicocelectomy can be advocated in men with oxidative stress. Until these studies become available, selective ligation of grade II/III varicoceles in men with poor reproductive outcomes despite oral antioxidant therapy is probably reasonable practice.

Vitamin and antioxidant supplementation

Elevated homocysteine has been linked with oxidative stress. **The B group vitamins folate, Vitamin B_6 and Vitamin B_{12}** are known to increase the enzymatic efficiency of the MTHFR and cystathionine β-synthase enzymes responsible for removing homocysteine from the circulation (Matthews, 2002). While yet to be proven to enhance sperm quality, the use of a B group vitamin supplement (5 mg folate, 100 mg Vitamin B_6 and 100 µg Vitamin B_{12}) is probably warranted in any man found to have hyper-homocysteinaemia and oxidative stress as this treatment is inexpensive and without significant side effects.

To date, over 30 studies have been published examining the effect of various antioxidant treatments on sperm parameters and pregnancy outcome. With such a large body of evidence it would be expected that firm conclusions regarding the clinical effectiveness of oral antioxidants on sperm function and pregnancy outcome would be available. Unfortunately this is not the case because of the use of different types and doses of antioxidants, lack of proper prospective placebo controlled study design and small sample sizes. Many small non-controlled trials report significant improvements in sperm count, motility and morphology while on antioxidant therapy (reviewed in Agarwal *et al*., 2004). However, as these studies are open to bias this review will only consider properly conducted placebo controlled trials or prospective trials measuring oxidative stress end points (sperm peroxidation and DNA damage).

Several studies have reported that levels of ROS within semen can be reduced by augmenting the scavenging capacity of seminal plasma using oral antioxidant supplements. The oral antioxidant Astaxanthin (Comhaire *et al*., 2005), carnitine (Vicari and Calogero, 2001) or a combination of antioxidants such as acetylcysteine, β-carotene, Vitamin E and essential fatty acids (Comhaire *et al*., 2000) have all been

shown to directly reduce seminal ROS levels. A randomized control study comparing 3 months of Vitamin E (600 mg/day) treatment with placebo has confirmed this reduction in seminal ROS levels (Kessopoulou *et al*., 1995).

Furthermore, a combination of **400 mg of Vitamin E and 225 µg of selenium** (Keskes-Ammar *et al*., 2003) or 300 mg of Vitamin E alone (Suleiman *et al*., 1996) have been shown in placebo controlled studies to reduce sperm MDA levels. Finally, a well-designed RCT of 2 months treatment with **1 g of Vitamin C and Vitamin E** reported a very significant reduction in sperm DNA damage (Greco *et al*., 2005a, b). This finding is supported by non-controlled studies which have also reported a reduction in sperm DNA damage with the use of a combination of **Vitamin C and E (400 mg each), β-carotene (18 mg), zinc and selenium** (Menezo *et al*., 2007) or a combination of acetylcysteine, 180 mg Vitamin E, 30 mg β-carotene and essential fatty acids (Comhaire *et al*., 2000).

While many relatively poorly designed studies have shown antioxidant supplements to boost sperm count and morphology, the majority of good-quality studies do not (Agarwal *et al*., 2004). The only parameter that appears to be possibly improved with oral antioxidant therapy is sperm motility. Many well-conducted studies have shown small but significant improvements in sperm motility with supplementation of carnitine (Lenzi *et al*., 2004; Balercia *et al*., 2005), selenium (Scott *et al*., 1998), Vitamin E (Suleiman *et al*., 1996), Vitamin E and selenium (Keskes-Ammar *et al*., 2003), glutathione (Lenzi *et al*., 1993) and Astaxanthin (Comhaire *et al*., 2005). However, two prospective RCT comparing Vitamin C and E supplementation with placebo have found antioxidants to have no ability to improve sperm motility (Rolf *et al*., 1999; Greco *et al*., 2005a).

While many studies have show improvements in sperm quality with antioxidant treatment, the ability of these changes to translate into improved chances of pregnancy is less clear. Suleiman *et al*. (1996) reported that treatment with Vitamin E resulted in a significant fall in ROS damage to sperm and an improvement in spontaneous pregnancy rates during the next 6 months (21% pregnant rate in the Vitamin E group V 0% placebo). Conversely, Rolf *et al*. (1999) did not report any improvement in spontaneous pregnancy outcome from 2 months treatment with a combination of Vitamin C and Vitamin E.

Finally, a recent RCT comparing the antioxidant formulation Menevit with placebo reported a significant increase in clinical pregnancy rate if the antioxidant was taken for 3 months prior to IVF-ICSI treatment (Tremellen *et al*., 2007). The Menevit nutraceutical is postulated to improve sperm quality by three complimentary mechanisms. First, it contains traditional antioxidants such as Vitamins C and E, selenium and lycopene to protect sperm from ROS already produced. Second, it contains garlic, which is known to have an anti-inflammatory effect, thereby potentially reducing seminal leukocyte ROS production (Hodge *et al*., 2002; Chang *et al*., 2005). Finally, Menevit contains zinc, selenium and folate that are believed to play a role in augmenting protamine packaging of sperm DNA (Kvist *et al*., 1987; Pfeifer *et al*., 2001), helping to protect sperm from ROS attack. While it is yet to be proven that combinational therapy such as Menevit improves sperm DNA integrity, it appears logical that using several antioxidants with different modes of action, together with an agent to reduce leukocyte ROS production (Vicari *et al*., 2002; Gambera *et al*., 2007; Tremellen *et al*., 2007) is most likely to result in a beneficial effect.

(Heather's note – please be aware if a pill put together with only the apparent active ingredients made by a drug company is found to improve sperm, taking better and more extensive products – and changing lifestyle factors can only do a better job).

Surgical extraction of sperm

It has been suggested that while sperm are in contact with Sertoli cells they are relatively protected from oxidative attack (Greco *et al*., 2005b), with most ROS-mediated damage occurring during storage in the epididymis (Greco *et al*., 2005b). Two studies have compared sperm DNA quality in the same individual using either ejaculate (Greco *et al*., 2005a, b) or surgically aspirated epididymal sperm (O'Connell *et al*., 2002) with sperm surgically extracted from the testicle. Both of these studies report significant improvements in sperm DNA quality in the testicle derived samples. Unfortunately neither of these studies assessed oxidative damage to sperm so it is presently uncertain if protection from epididymal oxidative stress is the sole reason for the observed improvements in DNA quality. As such, resort to the use of testicular derived sperm in men with poor DNA quality should only occur if more conservative treatments such as lifestyle modification and antioxidant therapy have failed.

Laboratory techniques to reduce the effects of oxidative stress

Centrifugation of a semen sample prior to its use in IUI or IVF can exacerbate sperm oxidative stress. This can be limited by reducing the time that the semen is centrifuged (Shekarriz *et al*., 1995a, b), use of non-centrifuge separation techniques such as 'swim-up' or glass-wool filtration and limiting the time in which sperm are cultured in media away from seminal plasma. Furthermore, culturing sperm under low oxygen tension (5%O_2/95% CO_2 versus 20% atmospheric O_2 content) has been shown to significantly improve sperm quality by reducing seminal leukocyte ROS production (Griveau and Le Lannou, 1997; Whittington and Ford, 1998). Avoiding use of cryopreserved sperm for fertilization is also ideal since ROS are produced during freezing and thawing of the sperm, thereby decreasing sperm quality (Watson, 2000).

Sperm preparation media may also be supplemented with a variety of antioxidants to guard against oxidative stress. The addition of catalase/SOD (Rossi *et al*., 2001), Vitamin C (Donnelly *et al*., 1999), Vitamin E (Donnelly *et al*., 1999; Yenilmez *et al*., 2006), ferulic acid (Zheng and Zhang, 1997), EDTA (Gomez and Aitken, 1996; Gomez *et al*., 1996), glutathione/hypotaurine (Donnelly *et al*., 2000), albumin (Twigg *et al*., 1998) and *N*-acetyl-cysteine (Oeda *et al*., 1997) to sperm preparation media have all been shown to protect sperm from oxidative attack. At the present moment commercial sperm preparation media does not contain any antioxidants aside from albumin and amino acids. Optimized culture media for sperm is unfortunately lagging well behind the complex sequential media developed for embryos and certainly needs intensive research as soon as possible.

OVERVIEW

An expanding body of evidence now supports a role for oxidative stress as a significant cause of male infertility (summarized in Table III). However, despite being a common pathology in infertile men, oxidative stress is ignored by many infertility practitioners. The currently popular response of resorting to mechanical techniques such as IVF-ICSI in all cases of male factor infertility is unlikely to be 'best practice' since ROS damaged paternal DNA will result in poor quality blastocysts, less than

optimal pregnancy rates and an increase in miscarriage. Antioxidant supplements have now been shown in randomized placebo controlled studies to protect sperm from oxidative related DNA damage and to boost pregnancy rates. It may therefore be prudent to consider using antioxidants in all infertile men exhibiting oxidative stress. Presently, one-third of men in infertile relationships already take such therapies (Zini *et al.*, 2004), indicating patient acceptance of antioxidant supplementation in combination with traditional ART treatments. Of course, antioxidants should be offered in combination with changes in lifestyle such as avoiding toxins (cigarette smoke, pollutants, heavy metals) and excessive heat.

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Table III.

Summary of the evidence linking OS with male infertility.

While a role for oxidative stress in male infertility is now established, many unanswered questions still remain. First, there is a clear need to develop inexpensive assays to identify sperm oxidative stress that can be easily conducted in any andrology laboratory. Secondly, large RCTs are needed to confirm the effectiveness of surgical interventions (varicocelectomy, testicular biopsy) in the management of oxidative stress. Further research is also required to determine what combination and dose of antioxidant supplement provides sperm with maximal protection against oxidative stress. Finally, the development of new sperm culture media that can better protect sperm from the ravages of ROS damage is clearly required.

Alcohol use and ROS

http://pubs.niaaa.nih.gov/publications/arh27-4/277-284.htm

Defeng Wu, and Arthur I. Cederbaum,

(Heather's note -This may be too scientific for some – yet it explains in detail what oxidative stress is and does, and what ROS is. Although not mentioning fertility, strong parallels can be drawn. The same antioxidants are mentioned as below - glutathione, Vit E and Vit C.

Of note is that they are using rats and mice, which make their own Vit C unlike humans – hence when speaking of liver damage and alcohol as this is, h-guinea pigs would present a far more disastrous result - as like humans, they need it to detoxify and protect their livers).

Smoking/ROS and using anti oxidants

http://www.clevelandclinic.org/reproductiveresearchcenter/docs/agradoc174.pdf
(Heather's note - Article explaining in depth the role of pollution, exposure to lead, smoking and any forms of oxidative stress and low sperm quality. The enhancement Vit C, E and Glutathione all make to improving make the fertility.

http://www.repromed.com.au/custom/files/Oxidative%20stress%20and%20male%20infertility.pdf

(Heather's note) - A comprehensive look at many of the common chemicals and lifestyle choices we are being exposed to; with the decrease in male fertility. Linking the anti oxidants to improving the fertility outcome.

Oxidative stress and male infertility—a clinical perspective

Kelton Tremellen

Oxidative stress occurs when the production of potentially destructive reactive oxygen species (ROS) exceeds the bodies own natural antioxidant defenses, resulting in cellular damage. Oxidative stress is a common pathology seen in approximately half of all infertile men. ROS, defined as including oxygen ions, free radicals and peroxides are generated by sperm and seminal leukocytes within semen and produce infertility by two key mechanisms.

First, they damage the sperm membrane, decreasing sperm motility and its ability to fuse with the oocyte. Second, ROS can alter the sperm DNA, resulting in the passage of defective paternal DNA on to the conceptus. This review will provide an overview of oxidative biochemistry related to sperm health and will identify which men are most at risk of oxidative infertility.

Finally, the review will outline methods available for diagnosing oxidative stress and the various treatments available.

Mechanisms of male infertility: role of antioxidants.

Sheweita SA, Tilmisany AM, Al-Sawaf H.
http://www.ncbi.nlm.nih.gov/pubmed/16248841

Abstract

Defective sperm function is the most common cause of infertility, and until recently, was difficult to evaluate and treat. Mammalian spermatozoa membranes are rich in polyunsaturated fatty acids and are sensitive to oxygen induced damage mediated by lipid peroxidation. Hence, free radicals and reactive oxygen species [ROS] are associated with oxidative stress and are likely to play a number of significant and diverse roles in reproduction. The excessive generation of reactive oxygen species by abnormal spermatozoa and by contaminating leukocytes [leukocytospermia] has been identified as one of the few defined etiologies for male infertility.

Moreover, environmental factors, such as pesticides, exogenous estrogens, and heavy metals may negatively impact spermatogenesis since male sperm counts were declined. In addition, aging is also likely to further induce oxidative stress. Limited endogenous mechanisms exist to reverse these damages. In a normal situation, the seminal plasma contains antioxidant mechanisms, which are likely to quench these ROS and protect against any likely damage to spermatozoa.

However, during genitourinary infection/inflammation these antioxidant mechanisms may downplay and create a situation called oxidative stress. Assessment of such oxidative stress status [OSS] may help in the medical treatment of male infertility by suitable antioxidants. The cellular damage in the semen is a result of an improper balance between ROS generation and scavenging activities. Therefore, numerous antioxidants such as **vitamin C, vitamin E, glutathione, and coenzyme Q10,** have

proven beneficial effects in treating male infertility. A multi-faceted therapeutic approach to improve male fertility involves identifying harmful environmental and occupational risk factors, while correcting underlying nutritional imbalances to encourage optimal sperm production and function.

Use of VIt C for just about anything

Esp when under stress and to detox the body (Vit C is a liver metabolite).

http://www.doctoryourself.com/realstory.html

Oxidative stress and antioxidants in male infertility: a difficult balance

Ashok Agarwal, Sushil Anandh Prabakaran,
http://www.bioline.org.br/pdf?rm05001
(Heather's note - (role of smoking and male fertility probs)

Abstract
Infertility is one of the most stressful conditions amongst married couples. Male factor infertility is implicated in almost half of these cases. Recent advances in the field of reproductive medicine have focused the attention of many researchers to consider reactive oxygen species (ROS) as one of the mediators of infertility causing sperm dysfunction. Although, ROS is involved in many physiological functions of human spermatozoa, their excess production results in oxidative stress.

Mitochondria and sperm plasma membranes are the two locations of ROS production that involves complex enzyme systems such as creatine kinase an diaphorase. ROS causes damage to the spermatozoa DNA, resulting in increased apoptosis (programmed death) of these cells.

The production of ROS is greatly enhanced under the influence of various environmental and life style factors such as **pollution and smoking**. An effective scavenging system is essential to counteract the effects of ROS. Various endogenous antioxidants belonging to both enzymatic and non-enzymatic groups can remove the excess ROS and prevent oxidative stress. Since, ROS is essential for the normal sperm physiology, rationale use of **antioxidants is advocated.**

Clinical Relevance of Oxidative Stress in Male Factor Infertility:

Ashok Agarwal, Kartikeya Makker, Rakesh Sharma
http://ccf.org/reproductiveresearchcenter/docs/agradoc261.pdf
(Vit C again)

Introduction
Infertility is a major clinical problem, affecting people medically and psychosocially. Fifteen percent of all couples in the US are infertile and the male factor is responsible for 25% of these cases. Of the many causes of male infertility, oxidative stress (OS) has been attributed to affect the fertility status and thus, it has been studied extensively in recent years.

Spermatozoa, like any other aerobic cell is constantly facing the 'oxygen-paradox'.

Oxygen is essential to sustain life as physiological levels of reactive oxygen species (ROS) are necessary to maintain normal cell function. Conversely, its breakdown products such as ROS can prove to be detrimental to cell function and survival.

OS has also been implicated in the pathogenesis of many other human diseases such as atherosclerosis, cancer, diabetes, liver damage, rheumatoid arthritis, cataracts, AIDS, inflammatory bowel disease, Parkinson disease, motor neuron disease, and conditions associated with premature birth.

Physiological role of ROS in male reproductive system. Until recently, ROS was considered toxic exclusively to human spermatozoa. Substantial evidence exists to suggest that small amounts of ROS are necessary for spermatozoa to acquire fertilizing capabilities.
Low levels of ROS have been shown to be essential for fertilization, acrosome reaction, hyperactivation, motility, and capacitation. Co-incubation of spermatozoa with low concentrations of hydrogen peroxide has been shown to stimulate sperm capacitation, hyperactivation, acrosome reaction, and oocyte fusion. ROS such as nitric oxide (NO) and the superoxide anion have also shown to

Clinical Relevance of Oxidative Stress in Male Factor Infertility:

An Update

Ashok Agarwal, Kartikeya Makker, Rakesh Sharma

http://ccf.org/reproductiveresearchcenter/docs/agradoc261.pdf

Male factor has been considered a major contributory factor to infertility.
Along with the conventional causes for male infertility such as varicocele, cryptorchidism, infections, obstructive lesions, cystic fibrosis, trauma, and tumors, a new, yet important cause has been identified: oxidative stress. Oxidative stress (OS) is a result of the imbalance between reactive oxygen species (ROS) and antioxidants in the body, which can lead to sperm damage, deformity and eventually male infertility.

This involves peroxidative damage to sperm membrane and DNA fragmentation at both nuclear and mitochondrial levels. OS has been implicated as the major etiological factor leading to sperm DNA damage.

OS-induced DNA damage can lead to abnormalities in the offspring including childhood cancer and achondroplasia. In this article, we discuss the need of ROS in normal sperm physiology, the mechanism of production of ROS and its pathophysiology in relation to male reproductive system. The benefits of incorporating antioxidants in clinical and experimental settings have been enumerated. We also highlight the emerging concept of utilizing OS as a method of contraception and the potential problems associated with it.

Conclusion
In the last decade, there has been a phenomenal growth in our knowledge of male reproduction, sperm function and development of diagnostic tools and treatment modalities. In addition, our understanding of OS has given rise to several new treatment modalities, which are now being investigated for improving male infertility. Many new antioxidants are now available that can decrease OS and improve sperm

quality but a major concern in their usage is lack of scientific evidence of their effectiveness, which has led to their non-approval by FDA.

However, OS being only one of the many causes of male infertility, it is recommended that antioxidant therapy should be tried only in cases of increased OS or oxidative damage.

Sperm swim singly after vitamin C therapy

(The answer to sperm agglutination – especially if smoking)
Elizabeth Rasche González
http://jama.jamanetwork.com/article.aspx?articleid=386823

ABSTRACT

Swallow a gram of ascorbic acid daily and restore fertility in just four days? It sounds implausible, to say the least. But Earl B. Dawson, PhD—with colleagues William A. Harris and William J. McGanity, MD, Department of Obstetrics and Gynecology, University of Texas Medical Branch, Galveston—has recently shown that this may be possible in men with infertility secondary to nonspecific **sperm agglutination.**

At the recent meeting of the Federation of American Societies for Experimental Biology in Chicago, Dawson reported on a study of 35 male patients who could not impregnate their wives because more than 20% (mean, 37%) of their sperm clumped together, as demonstrated by microscopic studies of semen samples. The men's serum ascorbic acid levels averaged 0.2 mg/dL, which is considered borderline by the US National Nutrition Research Council, in contrast to a normal level of 0.6 to 0.8 mg/dL. Semen levels of the vitamin averaged 4.2 mg/dL.

The improvement of Sperm Parameters and Chromatin Quality by Vitamin C

Mangoli E, Pourentezari M, Anvari M, Talebi AR, Nahangi H
http://www.sciencepub.net/researcher/research0411/005_11401research0411_43_49.pdf

Abstract:
Antioxidants are the main defence factors against oxidative stress (ROS) induced by free radicals. **Vitamin C (vit.C) and vitamin E** is believed to be the primary component of the antioxidant system of the spermatozoa and are one of the major membrane protectants against ROS and LPO attack. There is a relationship between activity of these antioxidant and function of sperm. Vitamin C, which is belonging to non-enzymatic antioxidant is used as a supplemented drug to improve sperm quality in male infertility. Given the importance of sperm in reproductive and

Generation health other hand and harmful effects of oxidative stress the aim of this study was to evaluate the protective effects of vitamin C on sperm parameters and chromatin quality in mice. 14 adult male mice were divided equally into two groups .mice of group 1 served as control fed on basal diet, group 2 received basal diet and

vitamin C (10 mg/kg/daily, intraperitoneal) for 35 days. Finally, right tail of epididymis was cut in Ham's F10.

Released sperm were used to analyze number, motility, morphology (Pap-staining) and viability (eosin-Y staining) of the sperm and DNA integrity and chromatin condensation assessments were ready by standard cytochemical techniques including: (AOT): Acridine orange, (AB): Aniline Blue, (TB): Toluidine blue and chromomycin A3 (CMA3). In vitamin C mice, a significant Increase was found in sperm number, sperm viability and sperm morphology compared to control group. Also a significant increase was found in sperm TB (normal) and Ao (double-stranded DNA) in Vitamin C group compared to control group.

It was concluded, Vitamin C not only is able to improvement the sperm parameters but also increases sperm chromatin condensation and quality in mice.

I suggest that if you are having trouble becoming a dad, regardless of what you have been told about HER state of health and reproductive wellbeing - follow science – even if the medical specialists seem not to see the relevance.

Why would you NOT choose to change your habits to make better sperm, which in turn would lessen the chances of embryo failure or miscarriage?

Why would you NOT use more Vit C, E, Selenium and either Glutathione accelerators or include significant amounts of magnesium and sulphates into your body, to enhance the state of your health and that of your prospective children?

Regardless of your state of health and her menstrual cycle – both of you taking these supplements and following a sensible cleaner life plan will only help you feel better and make far better babies than you otherwise would have – a win-win solution if ever there was one.

Below you will find solutions. To many questions you had not thought to ask!

The answers are grounded in what works. No research needed – as back through time – what worked was done and what did not was lost.

'Old wives' tales' were what kept all going through the hard times - as they were the anecdotal notes from generations back of mothers and their mothers whose job it was to keep the homes fires burning and all within the house alive and well – regardless of the weather and the harvest.

2 – YOUR FOOD FACTORY

It is one thing to read various supplements help improve sperm quality. What about the question – 'can you absorb them' - either as supplements or through your diet? You may want to see your body as more than being a refuse pit. It Is not just a case of add all in you feel like eating, and out comes the waste at one end. This may be the idea you may get after reading all information on the supplements above.

Your belly sustains life. It is fuelled by what an acupuncturist sees as Yang Qi. This may not be the language you are used to – however – the Chinese way of medicine has been used for thousands of years. Its terms are found in the glossary at the back of this work.

If you go to the site www.HeatherSays.com you will find an extensive glossary across the first banner under 'Healthy Life'. The second fertility/pregnancy related banner is accessible through clicking in 'Apps' in the first one – and here you will find extensions of all in this text that you may wish to follow for clarification.

What was written down thousands of years ago in China is still relevant today, as it was in those earlier times in a very different, 'down to earth' existence in another culture and country. It is immediately applicable to your life and in reading this you can see how you can change to instantly affect changes in the behaviours that you think you need to do: especially taking 'antacid' medication for the upset gut.

Better answer – stop upsetting your gut!! Heal what damage you have created. Your sperm will be made with better ingredients, as finally you will have a body that works well. Farmers forever have known how to get the best out of their stock and their land – otherwise all would starve. The health of the soil and the environment were crucial. They still are. We can only live on good foods that are grown from clean water and air and soil.

Without supermarkets to find food in, and without jobs to source the money to buy foods that sustain us, we would be lost. Similarly, without our gut working well, we are not able to sustain life.

Many who have multiple issues in their life find this reflected in their health. They may take pills from a doctor who dispenses not solutions, but the equivalent of red tape to pop over the warning light on the car's dashboard. Is this the best you want for yourself? It is highly unlikely that your health is perfect and you are just not making great sperm. You are probably quite stressed and generally unwell also.

Humans are very able to adapt and within each generation there are changes to what runs the body – here I suggest you pay attention to biological needs to allow your gut to work, so you can make a better bunch of sperm to make much better babies.

How to change this – follow the owner's manual instructions!

3 - HOW A WELL BODY SUPPORTS LIFE

From an energy model, looking at the normal everyday process of food in, and waste out, is very different. This information may not be seen anywhere else - please stay with me here, as understanding the energy model grants you the tools to improve your life.

Seen from this model the 'food factory' works like this - food goes into the pot (your stomach) with the fire underneath (being the 'Spleen Yang'). Enough 'oomph'/juice /fuel, and the food is digested well. The energy that drives the body is not visible or mechanical, hence is ignored by orthodox medicine. Studying any cultures traditional medicine **why** things happen is crucial to getting to the source of helping changes and thus healing. Knowing what to do can save your a lot of grief.

Please remember that this is an energy explanation.

The pure aspects of the energy transformation process pass upwards and the more gross physical parts are sent in their downward path. As more nutrients are extracted, the relatively 'pure' Qi/ingredients go off into different directions – western medicine knows that physically the portal vein carries the nutrients extracted from the small intestine organ to the liver organ - a major correlation in physiology.

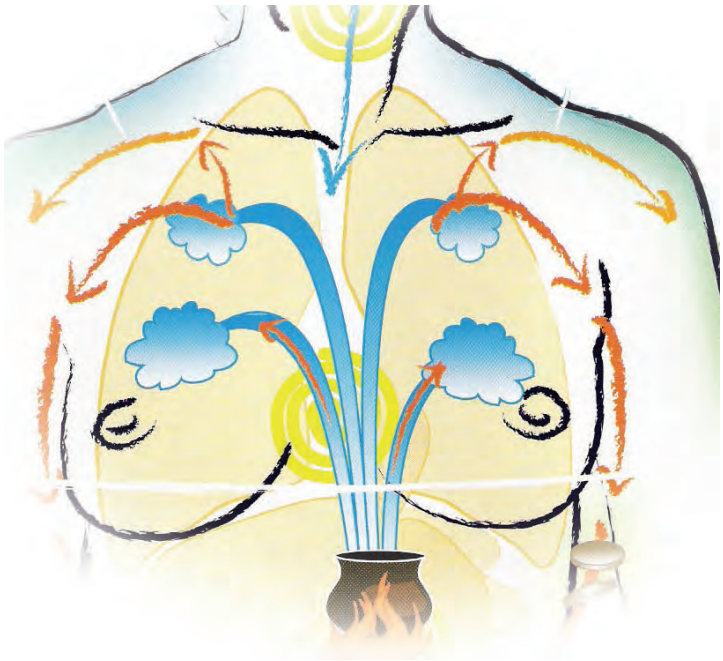
The Middle 'Heater' - where you would imagine it to happen. Food in, wastes out

The **golden spiral** is a representation of (from Indian traditions) a chakra – an energy centre. The **coins** – the precious reserve that if enough is left over from daily living is stored as Jing reserves – in the Kidney energy complex.

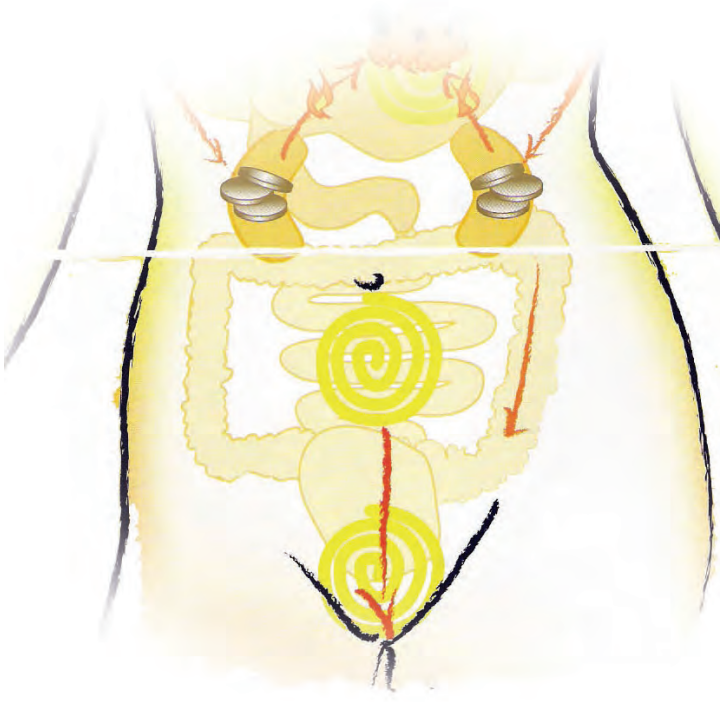
The Kidney Yang is also used as the heating used to digest the food – a catalyst - if we live well and moderately within limits. The pot is the 'stomach fire'. If fluid or food is too cold and put into the stomach, the fire may dwindle – and problems result.

These are felt as digestive complaints, allergies and nausea. Life suffers. Sperm are made from what is left over from running your own body. If you are not in a vital state, neither will be your sperm. The gut is the key – not just what goes in, but what it can do when the food is received.

The Upper Heater - The **blue lines** are the purest energy ascending into the lungs to start the energy circulation, which keeps the body flowing – which acupuncturists tap into along the meridians. The **red/orange arrows** represent the direction of flow.



The Lower Heater - Food residue descends. Once all the goodness is gone, the solid wastes are excreted. The fluid wastes from life processes are also discharged.



The **spirals** are representing chakras (energy centres); **coins** – what you are making as special reserves; sperm being one – how great your gut - how great your sperm.

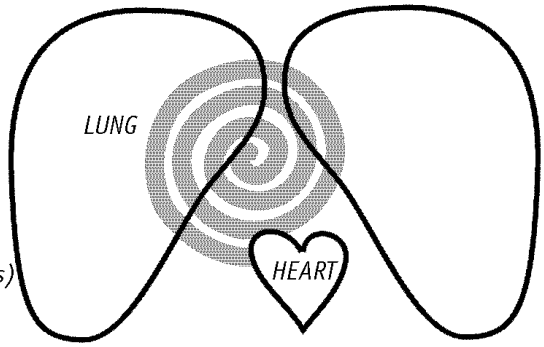
What is needed for the 'food factory' to work perfectly?

Energy model **vs** **What happens physically**

UPPER HEATER

RESONANCE

- *Heart chakra resonating as designed – absence of the consequences of uncleared grievances, shutting off own love*
- *Cosmic qi/prana*
- *Fresh air (charged with ions)*
- *Lungs free of grief*
- *Liver qi flowing, allowing the diaphram to expand properly*



SUBSTANCE

Presence of oxygen
Presence of pollutants
Breathing cycle free of all restrictions
Structure as designed to be - no spinal or organ misalignments

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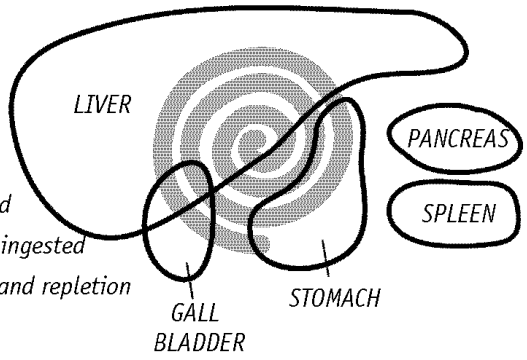
MIDDLE HEATER

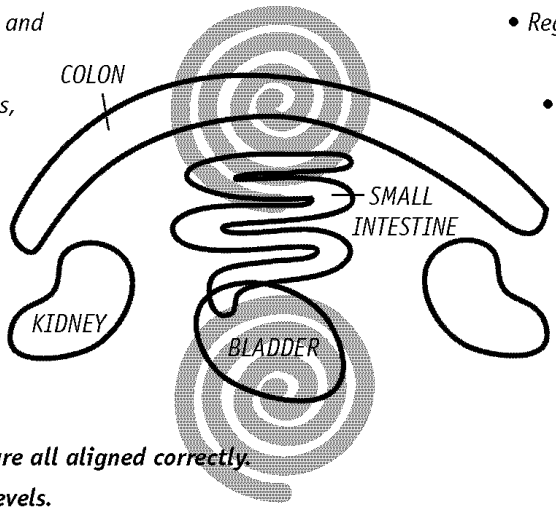
- *Solar plexus chakra resonating as designed Free flowing liver qi - if it is stuck, due to unsaid/unfelt/unexpressed emotions, will consequently totally disrupt all natural digestive flow*
- *Frequency, temperature and energy vibration of the food ingested*
- *Regular cycle of emptying and repletion*
- *Availability of yang qi*



- *Choice of quality, type and amount of flood/fluid*
- *Digestive enzymes appropriate to food requirements*
- *Backlog of toxins in the liver/gall bladder*
- *Presence of co-habitants - parasites and their toxins*
- *Posture and Structure*

LOWER HEATER

- *Presence of stuck qi, blood, cold, damp, obstructing the energy and eventually physical flows*
- *Presence of sexual blockages, fear, shock, rage, entrapped in the pelvic memories and energy fields*
- *Ability to feel/be here/ let go and experience*

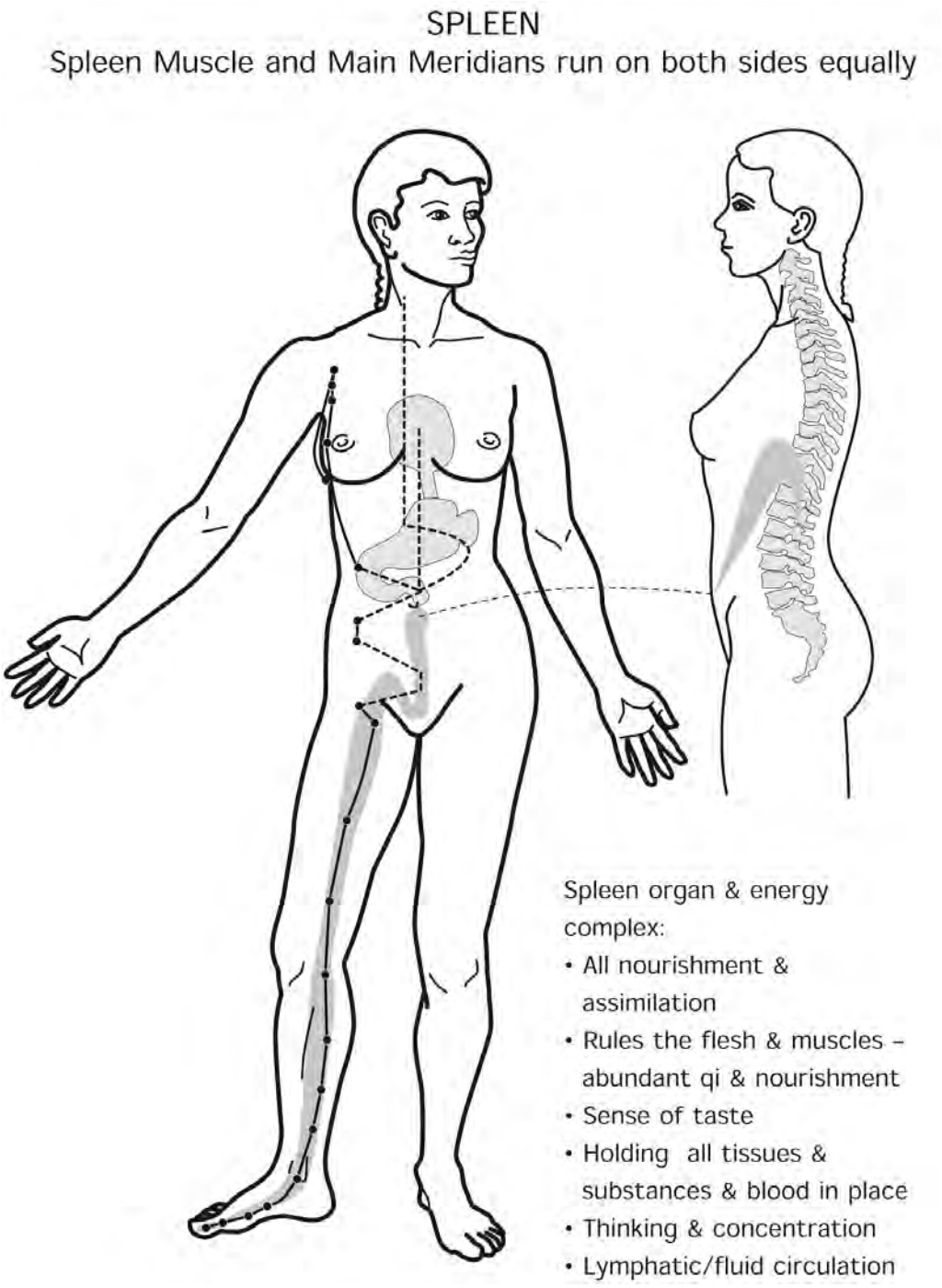


- *Appropriate waste disposal*
- *Regular appropriate sexual pleasuring*
- *Physical blockages as a result of the stuck qi*

Chakra, structure and posture all aligned correctly. Absence of scarring on all levels.

Next energy construct – acupuncture meridians

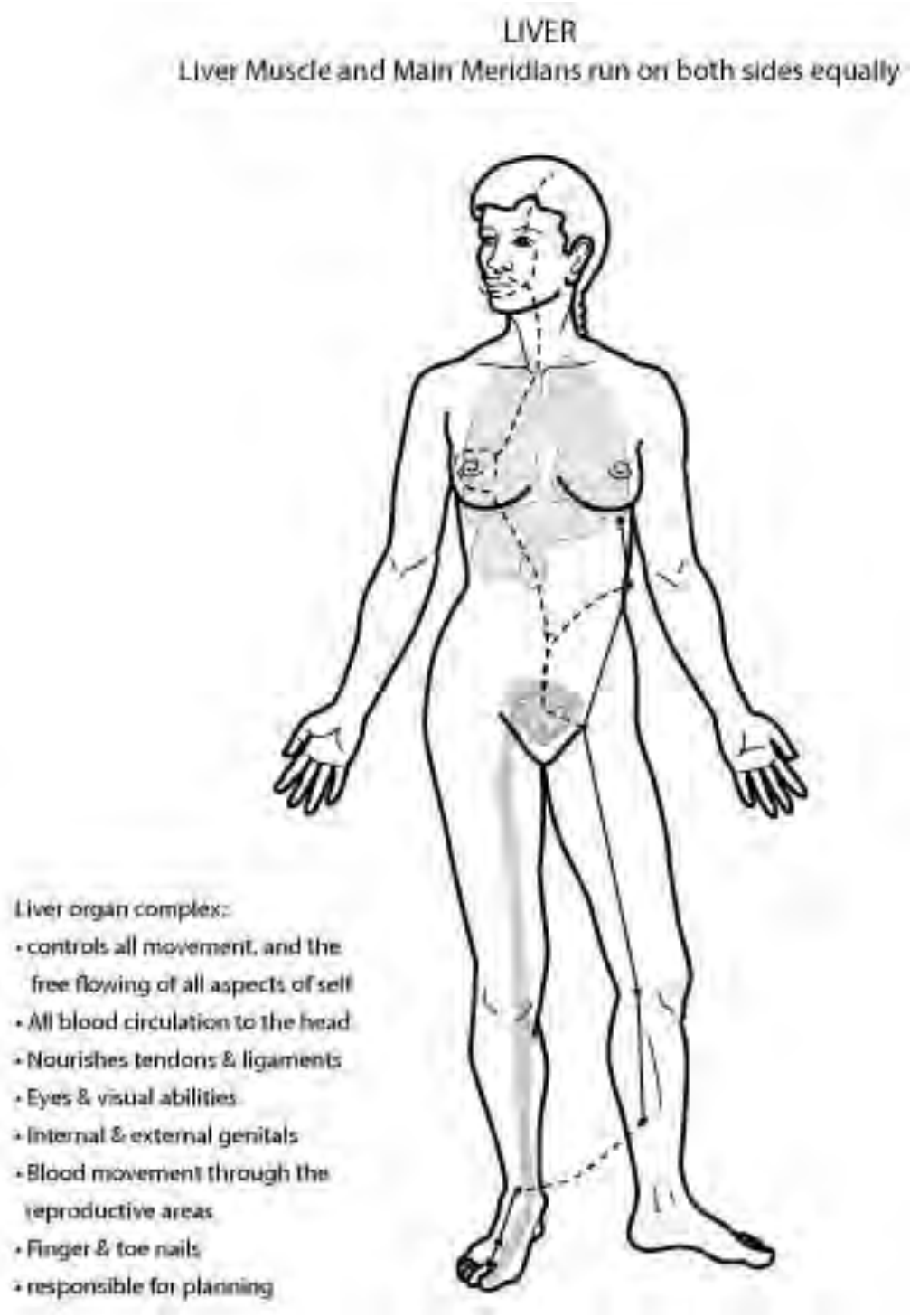
The entire body is serviced by energy grids - much like electrical circuits within a house, they are found all over - inside it, around it, on the surface - interrelated.



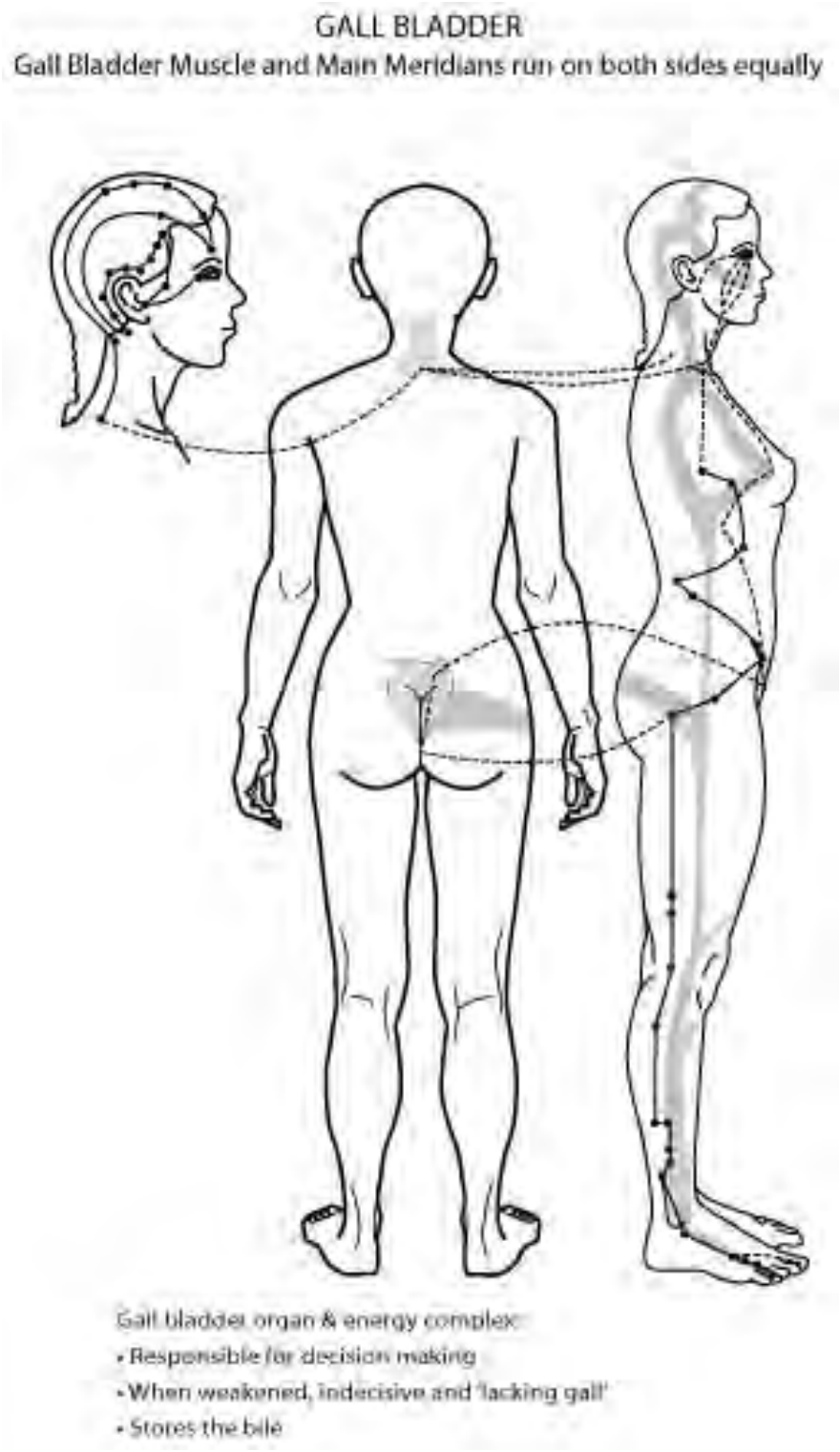
The Spleen meridian is controlled (as all are) by the amount and ability of the Qi to flow – itself a Yang Qi function. Scars, trauma and other blockages can stop up the circulation, creating local and distal problems, often decades later.

The presence of cold – locally lodged - can vastly upset this.

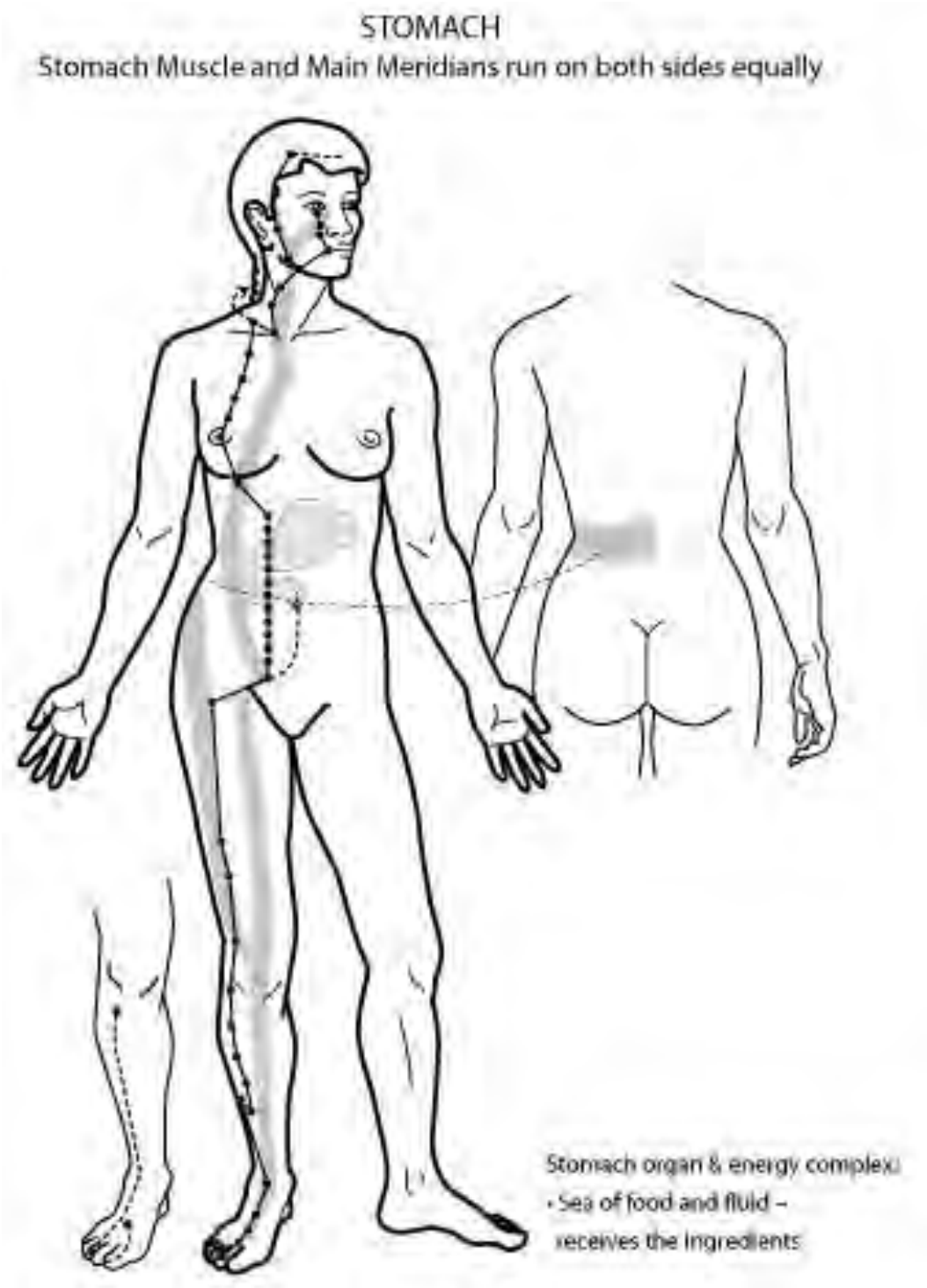
See all written in 'Cold – is NOT a Woman's Friend'.



The Liver meridian and Gall Bladder one work as a team – as such, anything that upsets your emotions, then plays out in the structural catastrophes that some live their lives through - simple answer – get more Magnesium oil on board to loosen up the tendons and the emotions! Good sources of magnesium?



Note where all the pain and tension and then need for structural people to realign you – try undoing your inner angst and using magnesium and you may be very pleasantly surprised – it is all just you being STUCK .



See how the meridians (they are all on both sides) cover so much of the area that is cut through in all operations - C sections, and breast surgeries/implants. Congested energy flow is often the beginning of the problems found later in functioning.

These can be easily addressed whilst visiting an acupuncturist – as using preventative medicine – not waiting till things fail and break down – is what you do with your car. Particularly so if you want great breastfeeding to happen – take steps to set this in place well before baby is on board.

How this model for digestion all fits together. The three heaters are as below –

Upper Heater – Breathing and circulation.

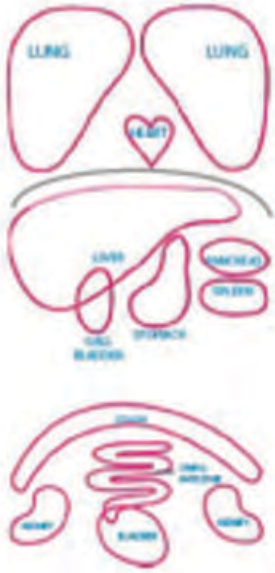
- Where the Shen (in the Heart) resides.
- Where all the Qi flow begins.

Middle Heater Physical Digestion

Receiving raw ingredients and apportioning perfectly.

Lower Heater – Uterus/fetal palace/Dan Tien – the energy centre of the body - Kidney home, where separation of the pure of the impure continues.

Relative placement of organs



In the **Upper Heater**, the most 'pure' resources are turned into the meridian Qi, after the pure resources from the stomach have mixed with the cosmic Qi and air from breathing. This is the grid that acupuncturists use.

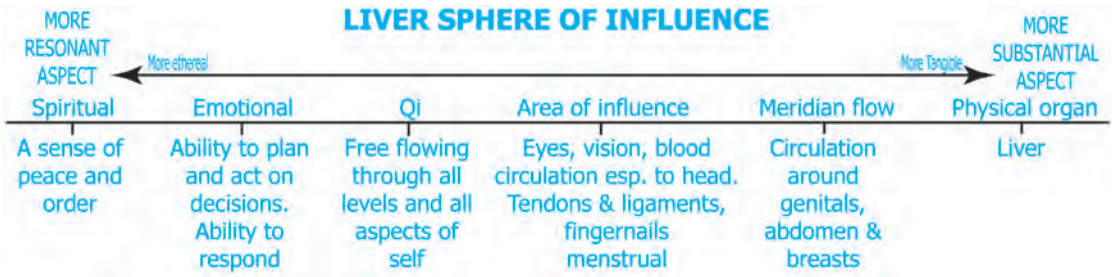
Middle Heater – where the food is received and the Spleen Qi./Yang transforms through 'rotting and ripening' (digestion) onto what is needed, and what is to be excreted is sent downwards. The purist physical components are sent across to the liver organ.

The **Lower Heater** – where the physical dregs go – and where the transformation into fluids for discharge and solid wastes happens in an orderly fashion. When there are 'left over' reserves from running the body, this Jing is sent to be stored. Here, the purest resources, alchemically transformed through digestion, rest in the Kidney complex.

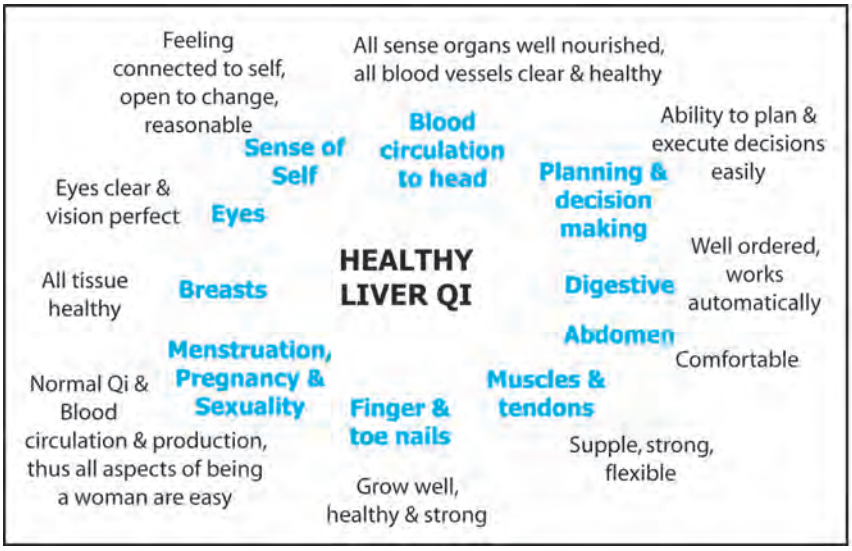
All this works perfectly with a few pre requisites.

1) - Liver Qi flowing freely. The key to all working in the body.

Liver QI needs to move.



What upsets the Liver Qi most? **Emotional constraint** – feeling and not expressing – anything – otherwise called *'stress'* in western culture. Resentment bottled, anger and unfairness festering. Over time it kills us. We block normal flow. Get bound up – growths and blockages and cancers - after years of internal dialogue and self attack.



The Liver Qi needs good Blood to nourish it – this is shows up the circular nature and the interdependence of the body – all need to be perfect for the whole to work.

When things brew – and when tempers fray and get stuck due to the behaviours we are modelled into (to be civil/nice and liked by all) - the energy has to go somewhere – and it does - one option is across and attack the gut. ('Upset tummy'. IBS, ulcers, heartburn, Krohn's disease, allergies, general gut malaise.)

It could boil over into tension felt in the structure, or by accepting the 'family' history of migraines, bad temperedness – especially when the Liver Qi is enhanced – and more Blood is in the body at period time – hence PMS. Most are aware that they are not happy – and just get used to it. BUT – the Stuck Liver Qi makes its presence felt.

All these different medical specialities come down to a basic cause – not happy

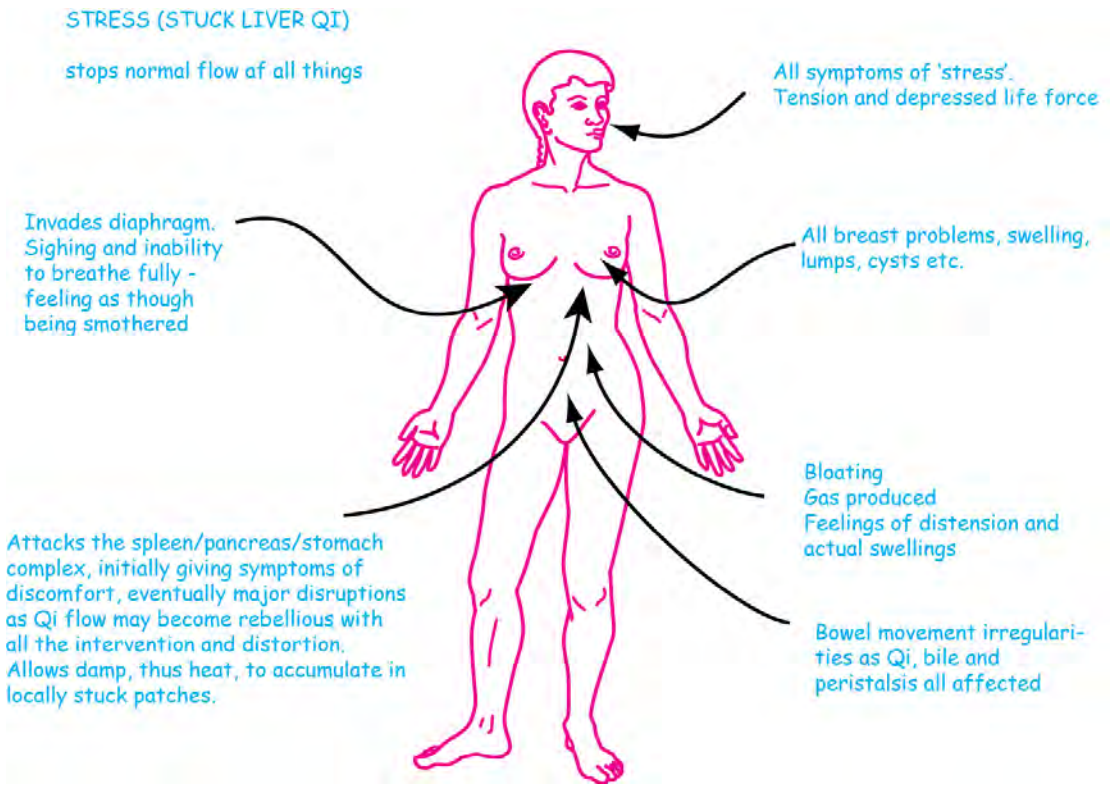
What can you do about this?

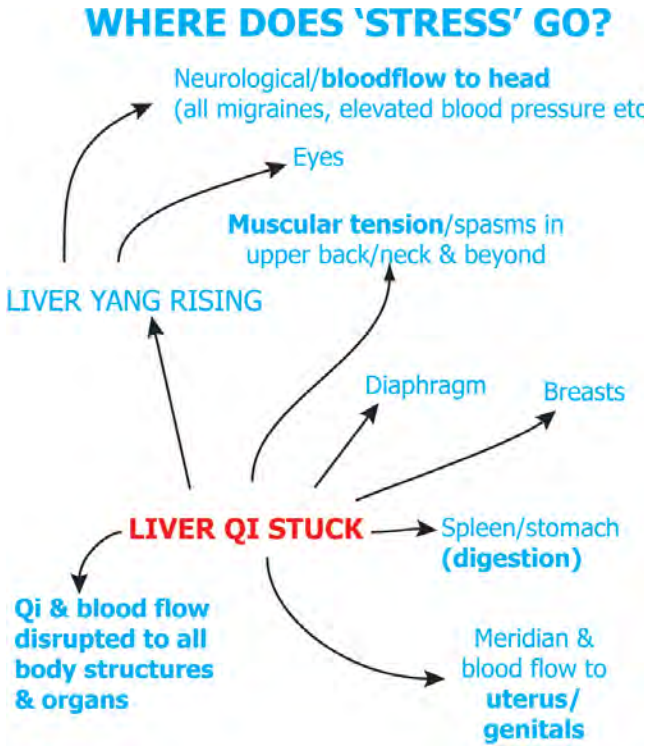
Look to all on the site www.HeatherSays.com - (e.g. moving Stuck Liver Qi) and the fertility app can guide you through my other informative sites – all for the further investment of just $2.

It does not have to cost in the usual sense. It may be as easy as starting an exercise regime. Singing, spending time with nature and starting up relating conversations with loved ones can only help how you feel. Stop inner festering improves all for you.

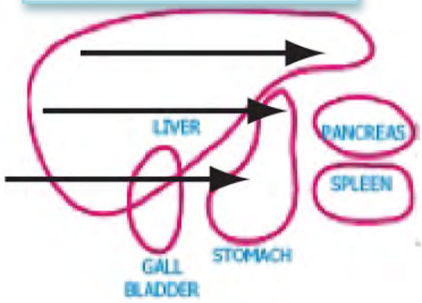
Using magnesium oil or spray externally at least 6 times daily on your skin will also improve how you feel and are.

The Spleen Qi gets wrecked (ulcers/IBS, heartburn etc) when you are annoyed – the Liver Qi invades the gut and there you have at least the beginnings of an inner volcano.





Look to where the Gall Bladder meridian flows – all through the 'tension' area that the chiropractors and massage therapists spend much time and your money trying to address.



Short answer – perhaps look to using Magnesium transdermally? (On the skin)

Why is this in a sperm enhancing book?

Because if you had all this working, there is no way you would need to read about it. The framework of damage control is not working for you – back to basics…and for your partner now, and when she is pregnant.

2) - Great Jing – from the parents and a moderate life to enable the 'leftovers' to accumulate so we have some for emergencies. (Also for when making babies). The entire system runs perfectly as long as there is sufficient Jing as the source substrate /foundation. (Here we have – you must fix this NOW as your precious baby is NOT getting all its birthright).

Are you BOTH well enough to be thinking of pregnancy?

3) – Proper ingredients – in moderation, variety, correct temperature, and harmony.

4) – Yang Qi able to work without stored cold wasting its ability to function (see Cold is Not A Woman's Friend). **If you are nauseated, or prone to vomiting;**

REASONS FOR NAUSEA/MORNING SICKNESS	ACTIONS TO COUNTERACT
1 -**Stomach Qi is too weak**/or empty (also Spleen Yang weak) →	1 } **Eat small amounts of easily digested foods, very often.**
2 - **Stomach Qi is too full**. →	2 }
3 - **Stomach Qi is too cold**.(also Spleen Yang is weak) →	3 -**Avoid all cold/chilled/raw foods and fluids**, especially when hungry/when stomach is empty, as then the Spleen Yang Qi is most vulnerable to cold penetration.
4 - **Stomach Qi is insulted/assaulted**. (over 'ripe'/damaged food). →	4 -**Avoid 'rubbish'/all toxicity/including odd mixtures/food combinations**. Avoid anything sweet in taste.
5 -**Stomach Qi is being attacked by Liver Qi Stuck** (stressed). →	5 - (Stress) **Recognise it**, and change especially eat in serene settings.
6 -**Abdomen is overfull** - Poo OR just that the developing uterus has a different qi flow, and is not yet being fully used by the baby - the body re-adjusts as pregnancy develops. →	6 -**Ensure daily, complete bowel movement**. In early pregnancy, the state of Mum's qi is often weak, and herbal tonics may help, also abdominal self-massage

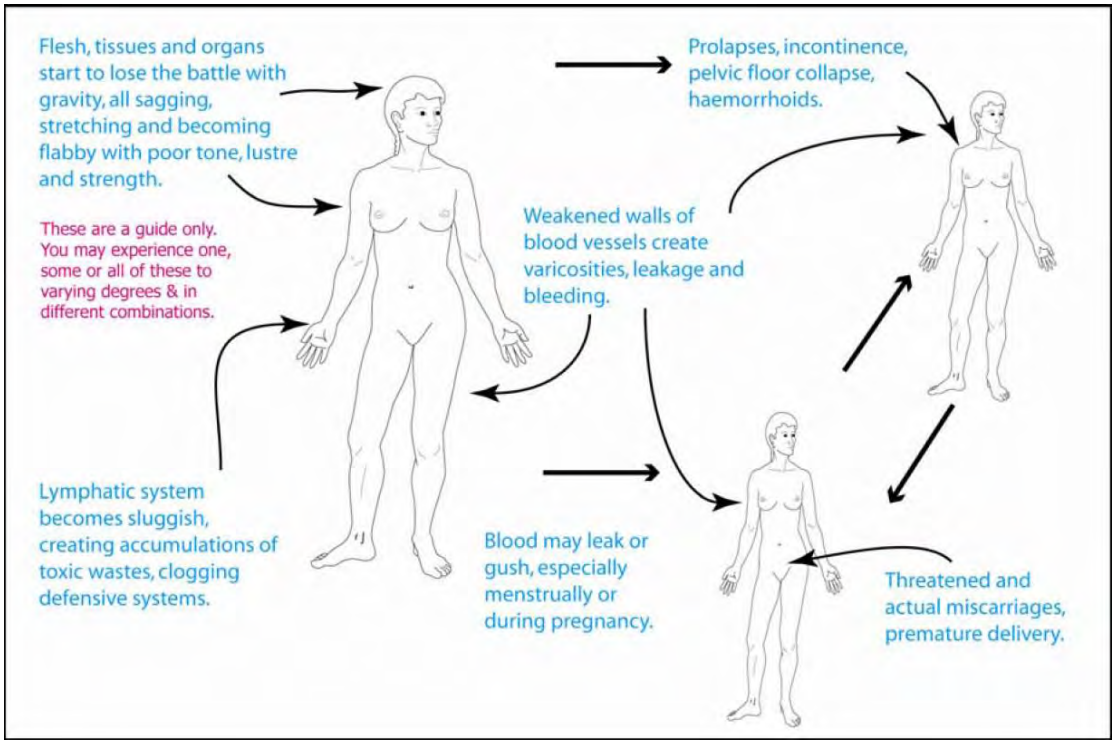
More is written on all these subjects in the fertility app that is so easily read through its categories 'The Problem', 'The Why' and 'The Solution'.

This is a ready reckoner for health – across all ages and stages - as it deals with how to improve yourself – for life and for baby making.

4 - DIGESTIVE TROUBLE

When the Spleen Qi/Yang can't work well, a whole raft of problems begin for you.

They may be considered 'normal' in life, or in aging, but are really just common, when the body starts to fail. When we do not look after ourselves – by essentially just doing what we feel like, regardless of consequences, the body starts breaking down.



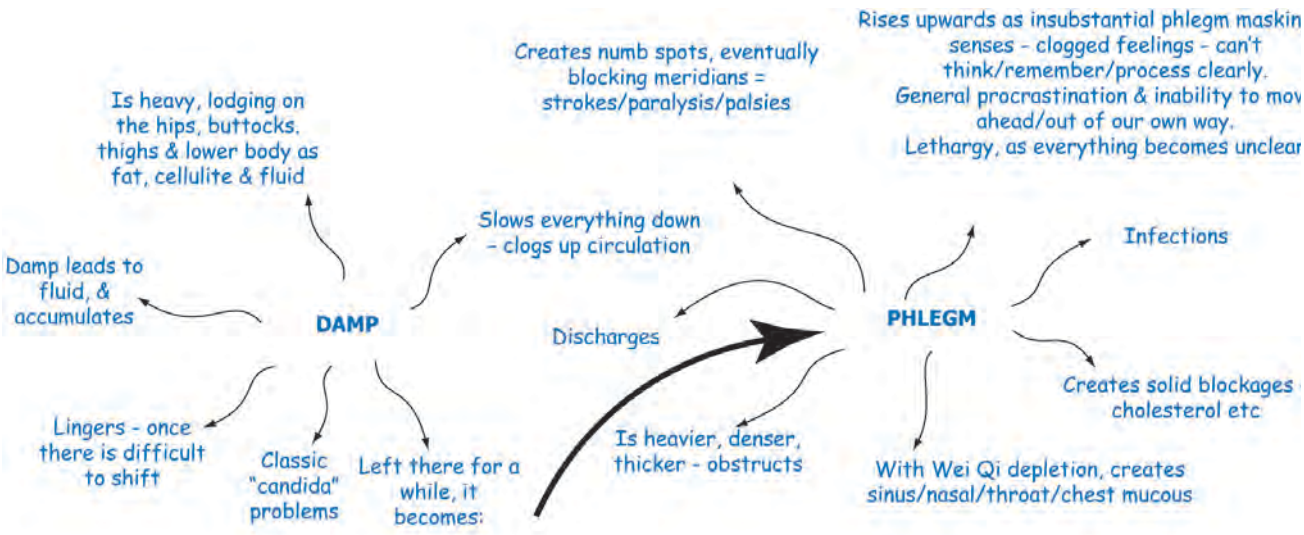
Dampness being formed is just one of the 'Spleen Qi deficient' problems

What do I mean by dampness? Anything that obstructs normal flow within the body – as the energy of dirtiness/fluid/heaviness. Often brought on through the weakening of the body's Yang Qi – and putting out the digestive fire. Exposure to cold (food/ fluids /environments) - eventually causes low immune/digestive/circulatory/thyroid function.

This is almost every man I now see in clinic to improve sperm.

It is an epidemic – whether from the recent exposure to WiFi and electromagnetic radiation from everywhere – not just wearing a mobile phone in a pocket that guarantees at least a third of all sperm is damaged – but all the exposure in minute amounts to myriad chemicals that were never supposed to be part of our makeup.

All of these mess with normal nutrient pathways. All of these may need to be removed in order for what you eat to be processed as your body needs them. Normal thyroid (metabolism) function is not measured by blood test – hence I suggest that you take your rectal BBT – if it is less than 36.6/8 C you will need to amend your life to ensure you can make great sperm for an enhanced digestion and immune system.



It takes often decades to get to where this is a big problem. You can help yourself – or wallow in being the same as always ... choice to change it is in your hands. Feeling hopeless and helpless whilst following the medical profession, which is so behind in wellness enrichment, will not help - look to a model that works.

Of course every one is just too busy to stop and enjoy being in their lives – fast is not a happy making process. Where will you fit a baby into all this busyness? Good gut function (Spleen Yang) and Kidney Yang (sexual/sperm) make great babies.

STOP CREATING THE PROBLEM

- Stop all your unnecessary outside activities and instead look after yourself.
- Especially if you have a cool - to - touch belly, don't add to the body coldness – nothing to be less than body temperature when drunk/eaten, or exposed to.
- At night — no fans on — no breeze, and not be compromised just because you think that you are 'too hot'. Fix this inner heat.
- All contact with the ground must be shod - shoes on. Ice in drinking water, or very cold air conditioning weakens the inner fore that supports life.
- Review the 'upset digestion' diagram as life is supported by your gut health. Start becoming responsible — don't continuing doing what makes it worse.
- Find a good acupuncturist, as this is easy to reverse using energy medicine.
- Source a moxa stick, and follow instructions here .

Cold abdomen

If you NEVER have a warm abdomen: This is a sign that all Yang Qi is struggling. See signs of a distressed Spleen Qi on the following pages. If you did not have a translation of the body codes — there it is — **avoid all cold**.

This means never walk around on cold floors - especially not at night when off to the loo. Have rugs on the cold floors. The cold invasion can start from contact up through the soles of the feet and lodge in the uterus/prostate and belly and back.

Where does this come into consideration in the orthodox medical model?

It doesn't. Iatrogenesis – caused by medical 'help'. Don't put ice anywhere near you. Cold is Not Your Friend Cold causes the energy to stop flowing - hence pain.

Feel your belly with the inside of your arm — is it warm — does the temperature change between above and below the navel? The key factor in determining if you need to boost Spleen or Kidney Yang in your body will be obvious – just look at the symptoms below.

There used to be a cupping-cold-out-of-the-navel procedure that was done in all Chinese families before the modern Cultural Revolution and the recent push for modernisation. China, like the west, has had a tendency to throw away what used to work, in ancient times. In this case, there IS nothing from modernisation to replace it.

' Spleen Yang' deficiency vs 'Kidney Yang' deficiency

This may sound like a foreign language. It makes sense when you start reading it though – so please persevere. You will probably find yourself fall through the cracks of medical help, usually with a raft of seemingly minor complaints. See what you are doing to create these, and so what to stop doing, to relieve yourself. The symptoms listed in general Yang deficiency are a guide only. The key factor in determining if you need to boost Spleen or Kidney Yang in the body are obvious if the following indicators are present. (All body Yang originates from the Kidney Yang).

Using other terms that are more western-friendly just does not work here. 'Spleen' is more than just an organ function, although it includes that of the pancreas. It is more about the digestive function, and the lymphatic, and the holding blood and tissues in place. When Spleen **energy** is strong, the digestive system (as everything) just runs on automatic. Just as when the Blood **energy** is strong, it allows all reproductive aspects, and nourishing all aspects of self, to be uneventful.

What damages this? Eating irregularly, and/or eating too many sweet or raw or cold things and/or also worrying and obsessing about life or little bits of it will waste the Yang Qi. Cold exposure is the largest influence. Unfortunately cold is often given medically as a treatment. Whilst icing a bit – like a ankle - may make it temporarily ***feel*** better, the whole raft of life quality issues may start sinking. This is often not noticed till well after the event and only then if the observer has been trained in a wellness model of medicine. Want more sexual fire? Don't use ice on anything.

SPLEEN YANG DEFICIENCY SYMPTOMS

Poor appetite. Without a healthy interest in food, the Blood energy cannot flourish, and life may be weakened/ compromised.

Loose stools/apparent diarrhoea. This may seem normal to someone who has been ill/not well for a while; but having unformed stool is a sign that the Spleen Qi is

not doing its job of transforming well. Undigested food there is even more of a warning sign. Any gut problem is likely to be better serviced by finding a practitioner who works WITH not in spite of, the body's natural flows. Great digestion and assimilation, make quality Blood and Qi and Jing, and then translate into both health and great sperm production.

Fatigue, especially after eating. The weakened Spleen Qi may be only capable of doing one thing well. Stopping all external activity at least ensures that there is energy available for the food to be processed. Brain fog is often a matter of low iodine and can be easily seen as a low thyroid marker.

Abdominal distension, gurgling, possibly discomfort and dull pain, especially after eating, show signs of weakening digestion and gut/hence sperm deterioration.

These imbalances are very serious if left untreated. Medical advice is only ever 'rest', and hope everything resolves itself. This is a little like having a broken leg 300 years ago — it would depend on the luck of circumstances as to whether you lived and had a leg that worked at the end of the waiting. Similarly, waiting for nature to right itself is only offered here as there is nothing MEDICAL (pharmaceutical/surgical) that is seen to help. This does not mean you need to sit it out – just change what created it.

What to do

- NOTHING cold is to be eaten/drunk, or used anywhere near you. The extra cold intake wastes what little metabolic energy (Yang Qi) you may have.
- Eating a little, cooked and nutritious snacks will assist the gut to rebuild.
- Using digestive enzymes with a meal may be a great help to digestion.
- Taking a good probiotic product will assist the gut flora. Most of the immune system comes from this. This alone may make an astonishing change to the apparently impossible gut problems you may have been living with.

In addition — there may be aspects of the Kidney Yang deficiency as below or this may be interspersed with other warnings from other body distress pictures.

KIDNEY YANG DEFICIENCY SYMPTOMS

Needing to pee a lot, especially at night. This may be seen as being 'normal' as you age. It is a sign of poor Yang Qi.

Weak, or aching lower back. If very strongly deficient, the knees and legs may also feel unstable and vulnerable. Whilst you may have had this forever – and see this as being normal — it is actually the beginnings of an overall depletion of inner resources, hastening the ageing process. You may also have **cold intolerance**, and **poor circulation**, with **cold hands and feet** — although may feel super hot and even be a sweating presence in bed at night – this is due to inner toxins. This is also often a **low thyroid function sign.**

Lack of interest in life or in sex. This may become lessened feeling, even with direct stimulation. Sensual numbness may set in, as the body shuts down non-

essential (for physical survival) drains on its most precious essence and Kidney Qi. Men do suffer from this – and it is a real concern to feel drained after ejaculation.

Again — low thyroid function is an unrecognised epidemic as the blood test numbers are unable to assess what is REALLY going on for the patient population. Best to invest in the book Thyroid Power by Richard and Karalee Shames. Also read Relationship thyroid to yang qi

What to do

Whilst it may appear obvious, it is important to ensure that you are sustaining life well with a great diet – and not eating ice/ice cream /drinking favourite fluids out of the fridge — heaping even more cold into your system.

Going back to what is the body SUPPOSED to do here – will help.

General yang building/tonification: Moxa

The energy model from acupuncture is included here, as there IS no answer in the mainstream thinking, past waiting for it all to go away, or using a new to nature/ with someone making lots of money from you-being-sick drug. Instead you can study whether it is the Spleen Yang or the Kidney Yang that is suffering more. It may be both, and if you are not sure, do everything I suggest below.

You may notice that a difference in the severity and the presence of cold for the more serious Kidney Yang deficiency type. Most of us, regardless of how well we think that we are, and how well we look after ourselves, find that ourselves here somewhat.

Whether it is through life – that is exposure to temporary climatic change, daily experiences, or the ministrations of various medical professionals, imbalances show as these symptom pictures. You may find results by finding an acupuncturist who also may be a Chinese herbalist using a wellness model gives quick results.

A stick of moxa may be obtained from most Chinese shops, or a local acupuncturist. You want to get the smelly/smoky one as the smokeless variety does not do the job. It is NOT just a matter of heat, but real vibrational herbal/energy medicine.

All use of moxa is to be performed where you are warm, without draughts, but where adequate ventilation is possible. See the Vimeo footage of the sacral fan process here. Make sure both of you are not hungry, and have drunk a glass of non-chilled water before, and after the process, regardless of thirst. This is a very heating and drying treatment. Whilst the smell lingers, and it is a hassle to go outdoors, and not get in a draught whilst doing this, using moxa has a profound effect on your health and wellbeing, if used as directed.

Please read the other eBooks in this men's vitality package as the messages there are to alter all of your life – not just 'fix' what has stopped you in your tracks. Often it is all about the inner story – the life that has impacted upon you – often due to early life, well before you had any say. As it appears to be not relevant to the physical workings of a body, no one else may mention this. So much of this debate centres around nutritional deficiencies allowing distress.

If you have attended to all mentioned in the physical realms - the gut flora is normalised, and the cold exposure is stopped and the right (temperature, variety and frequency) food is eaten and all is as it should be. The Human Microbiome is a great place to start reasoning through how to support great gut function.

Another major component is then stress and its effect in the normal workings of a body. I have covered this in depth in other writings that you can access as part of the Everything package.

Cold exposure wastes the Yang Qi, and ANY icing of injuries, drinking chilled water, eating cold foods especially on an empty stomach, and being in air conditioning, having fans on or windows directly open when sleeping all waste the Yang that is needed also to run your body well and keep the sexual fires topped up and your sperm in great swimming condition.

It may seem unusual to think of the programmes/energy running the physical body, but that is where the changes may need to be. Ignoring this aspect may be why everything you have done so diligently to date has not worked.

Without checking your BBT (Basal Body Temperature) you may never know that it was you all the time creating the miscarriage and pregnancy mishaps – get the thyroid within normal limits - as evidenced through the normal RECTAL BBT of 36.6/8 C for men (on arising after at least 4 hours straight sleeping).

You can view a presentation on this very topic here.

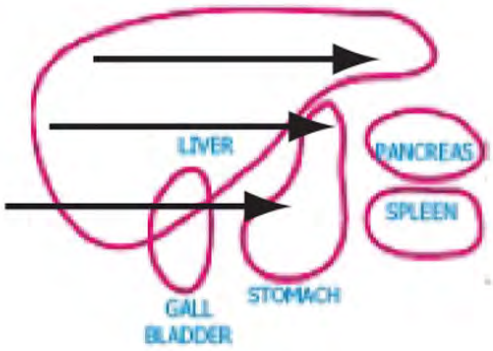
5 - SOLUTIONS

Sperm is about movement and action – and the best quality is found when the body is working optimally. It all is dependent upon the gut and its strength. Great Yang Qi.

Answers to upset digestion, (thus not great 'leftovers' to make sperm) are to be found here. Digestive energy (dependent on the quality and availability of your Yang Qi) is supposed to perform automatically without your having any discomfort.

'Stress' – 'Stress' is simply you're not moving through what has bothered you – to a new place of being totally present. This can set up your gut just not being able to work – and your reaching for antacid medication which will totally mess with your sperm. (This may result in wrong gut bacteria, the resultant Candida, and the eating patterns that keep them on line).

What sets up your gut distress .



Liver energy invading (from your being angry/ frustrated/ irritated/and bottling it, festering and keeping your peace /being 'civil' – all create the inner war where the Liver Qi goes across and attacks your gut. You may have all the western medical labels. After the digestive weakness, and before you actually resolve what causes it – the 'stress' of being you – you can get phenomenally ill – after the reflux, colitis, IBS and whatnot – through to ulcers and Krohn's disease and into cancers. See Liver meridian flow

(being 'nice'/ compliant/'easy to get on with').

Where does all that rage/fury reside? In your body – trapped there awaiting release - when too 'full' – it starts messing with life – think of a fridge or cupboard that is constantly being added to with nothing taken out /thrown away/used . Things fester too: eventually an overflow happens - as you may find in yourself. . It may be time to unload, before the pH is not right, allowing all gut bug invaders free rein.

What to do?

There will be clues in your 'story' - the one that you carry about with you and bring out all the time – your defining signature. What you have been calling what is happening to you 'normal', quite possibly no one else would . . .

Work through all in the Live Well eBook series. Stop breaking your body's blueprint – when you feel something – EXPRESS yourself. A well adjusted body needs to be working on 'automatic' without all the emotional charges and loops disrupting the regular flows that ensure wellness — moderated panic and/or shock and/or rage and/or terror all have a part to play in the average person's residue after making it to adulthood.

There usually needs to be a change if healing is to happen. Wanting to be a parent, yet continuing to do what you have always done will probably net you more of the same - not so good sperm. Reproductive disasters.

- **Work on what is really the problem** — it is the way to get to see you to start really healing (your life).
- **Stop all excess inner (thinking/worrying/obsessing) and outer activity.** (Going to work/keeping up appearances)
- **Exercise, meditation** and **yoga** are also likely to settle your nervous system.
- On the more mundane and obvious front — working with the gut directly.
- Often **just squeezing half a lemon into some warm water and drinking this prior to arising** can help with the physical liver/gallbladder's role of getting the peristalsis moving and the excess wastes discharged — before eating anything.
- Find **friendly gut bacteria** stored in refrigeration at a chemist or health food shop. Take as directed, to assist re-colonisation of the intestines, especially after taking antibiotics. Better still – eat fermented foods in your daily life.
- Take a **small dose of Vitamin C** frequently throughout the day, some liquid Zinc, and a good (possibly liquid) source of multi minerals, and vitamins.
- Pop magnesium oil on the skin any times a day – it will help all stress and gut and tension issues resolve simply why you might improve with magnesium
- A good soak in a magnesium (Epsom salts) bath may also relax you sufficiently to loosen the tension that has created the excess to sit within you.

Stressed person, stressed gut. Mal-absorption can become a hidden issue — you may be unaware that the good food consumed is not being used appropriately. Quality sperm rely on your digestion working perfectly.

This is covered in detail in '***The Livewell package: also the Men's Vitality package***

- If the belly — above or below the navel feels at all cold, especially if you still have cold hands and feet, or a cold bum at night — you probably need the cold removing and a Yang boost. (Here the moxa is wonderfully powerful).
- Sperm production is totally dependent on your having great Yang Qi.

There may also be a generalised weakness/aching in the lower back. This and the state of digestive weakness will depart when the Kidney and Spleen Yang are better.

Be aware that if you are eating or drinking anything cool/cold or raw or sweet you are helping to create the problem. If you are finishing a shower with cold water; if you are swimming in unheated pools; if you persist in having windows open or fans or air conditioners going at night when the Yang Qi is being replenished — you are helping to recreate your Yang Deficiency problems.

Think you are too hot? You are probably just too toxic, with a low BBT – read about inner heat in the Cold is Not Your Friend eBook. Start cleaning your insides out.

Health crises do not just happen — they are usually brewing for ages. The energy model from acupuncture theory allows you to see them gradually developing. Having a Yang deficiency has far reaching consequences, which are all avoidable. Spleen Yang Depletion.

What to do to fix this tendency for generalised weakening?

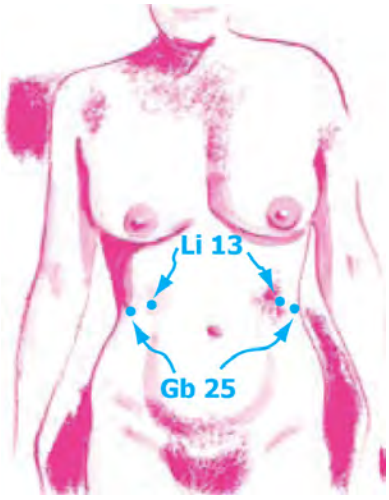
It is very possible that you can avert any energy drain, back-aching and weakness and frequent trips to the loo at night or in the daytime, plus improve your sex life AND your fertility, by ensuring that moxa is used as directed.

This is important also if there are sexual competency issues - and often premature ejaculation and lessened interest and desire are easily fixed (I am serious) just by using the moxa stick. I know that helping yourself may well be outside your comfort zone – but it is cheap, effective, safe and will also sort out any lower back issues – especially if going to the structural specialist is your high maintenance activity that just keeps happening - maybe all that is needed is your Kidney energy strengthened.

The quickest solution may be to find a holistic health care practitioner who uses a wellness model. Below may be too much info for some – but it does empower you to sort yourself out – and is totally safe and effective!! Improves sperm quality instantly!

Helping yourself

Is quick, easy and safe – and guaranteed to work. Instead of masking symptoms, or glossing over with drugs, you are actually getting to why things are not working.



Check yourself out - Points are found by pressing on the ends of the ribs — which may be a little hard to locate if well-padded. Lying on your back may make discovery easier.

If having trouble locating these, start at your back waist level and gradually move your hands around till you get to a free end of a rib: if it is the bottom one that is the point **GB 25**.

Moving upwards and towards the centre line a little, the next pointed rib will be the spot **Liver 13**. If either of these pairs or even one point are sore, there is a need to improve your body workings.

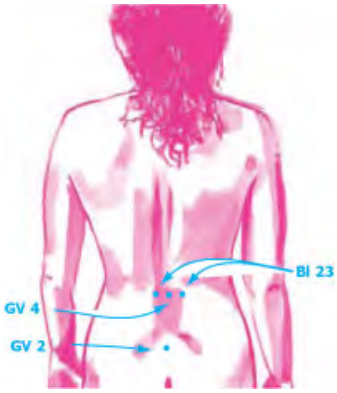
You will cause no damage by doing this – just fix what is the source of the problem – a weakened body. See which point is sorer - they may both be and equally.

The bottom point **(GB 25)** tells you the state of the **Kidney Yang.** This is the root of all activity in the body. Generalised yang deficiency causes such nuisance problems as having to get up at night to pee, with eventually occasional incontinence, often feeling too cold, not digesting food well, having circulatory problems and eventually sexual issues. No particular order – body does it all on its own timetable.

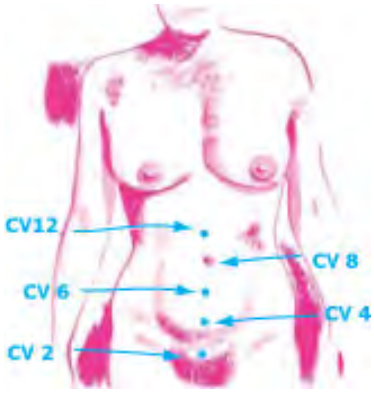
This may have been happening for a while - all aspects of the body start failing in a patchy, often random way.

You are feeling the end points of ribs - the next rib up (both are 'floating') is the point **Liv 13** tells you what is happening in the digestive/holding-the-blood-and-or-tissues-organ-body-in-genral-together-and-in-the-right-place - **Spleen Qi**).

You can help this by consuming only warm, nourishing foods, plenty of water, **no** caffeine, chocolate, alcohol, cigarettes /cool/ cold/raw foods or fluids, or sugar.



Using moxa over the points specified can be very soothing and also spur the body to work well again.



These problems due to exhaustion are so easy to shift. It is safe and easy and makes for far stronger sperm. While the energy reasons for the physical complaints are ignored, they grow, with the imbalances are worsening as time moves on.

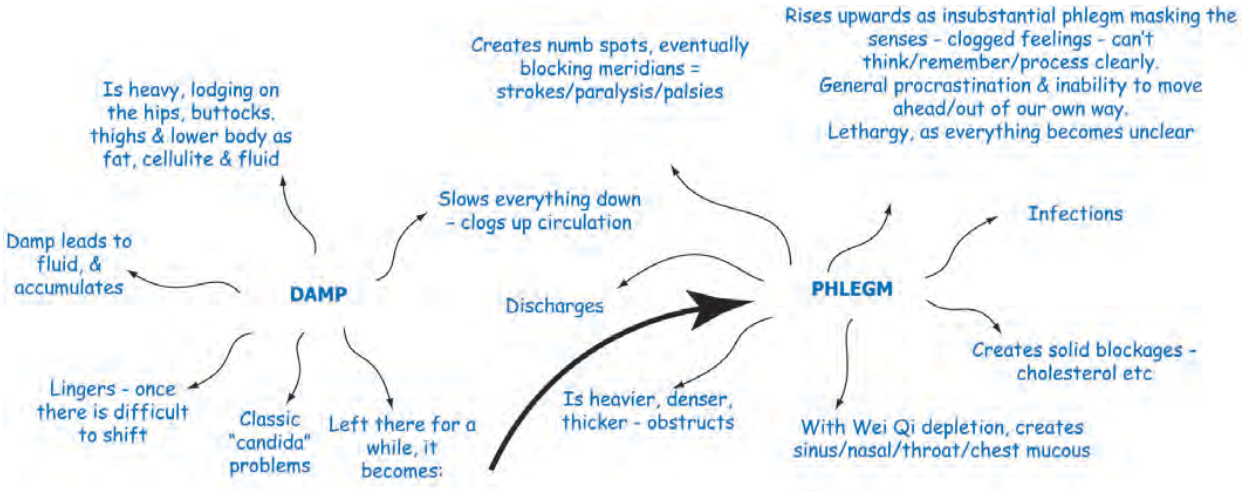
Exercise, go outside and sit in the sun - Vit D from sunshine is crucial for life.

Changing gear into becoming more of a human *BEING* and less of a human *DOING* may be of great help also.

When Spleen Qi is weak – a craving for sweet develops - it is as though the inner gremlins need feeding - and by doing so, you are well on your way for all manner of other problems. (Sugar – the Bitter Truth)

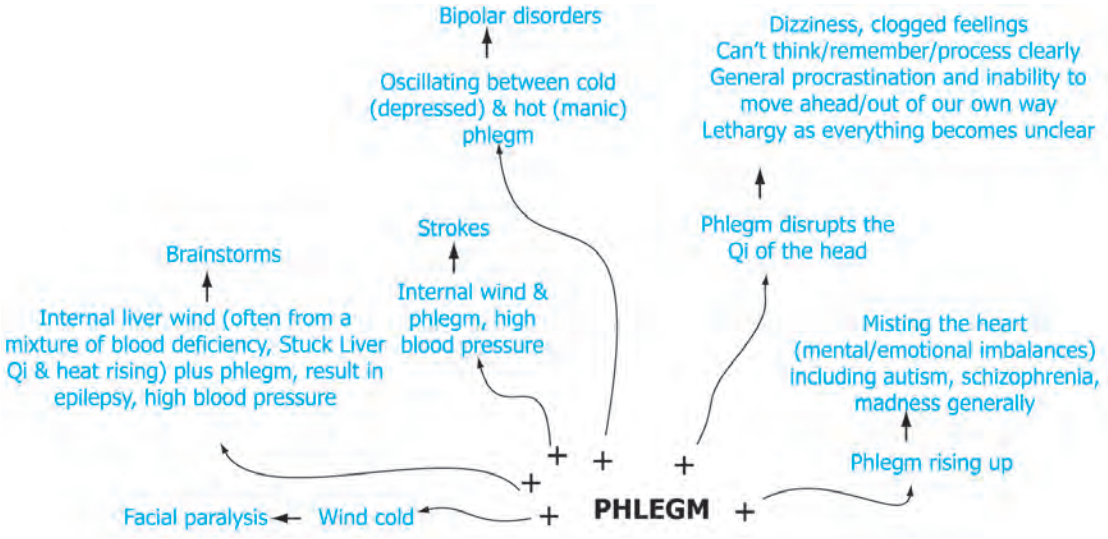
Damp leads onto **phlegm** – which is not just what you cough up from your lungs.

DAMPNESS? What do I mean? Sludge – the energy of obstruction/phlegm.

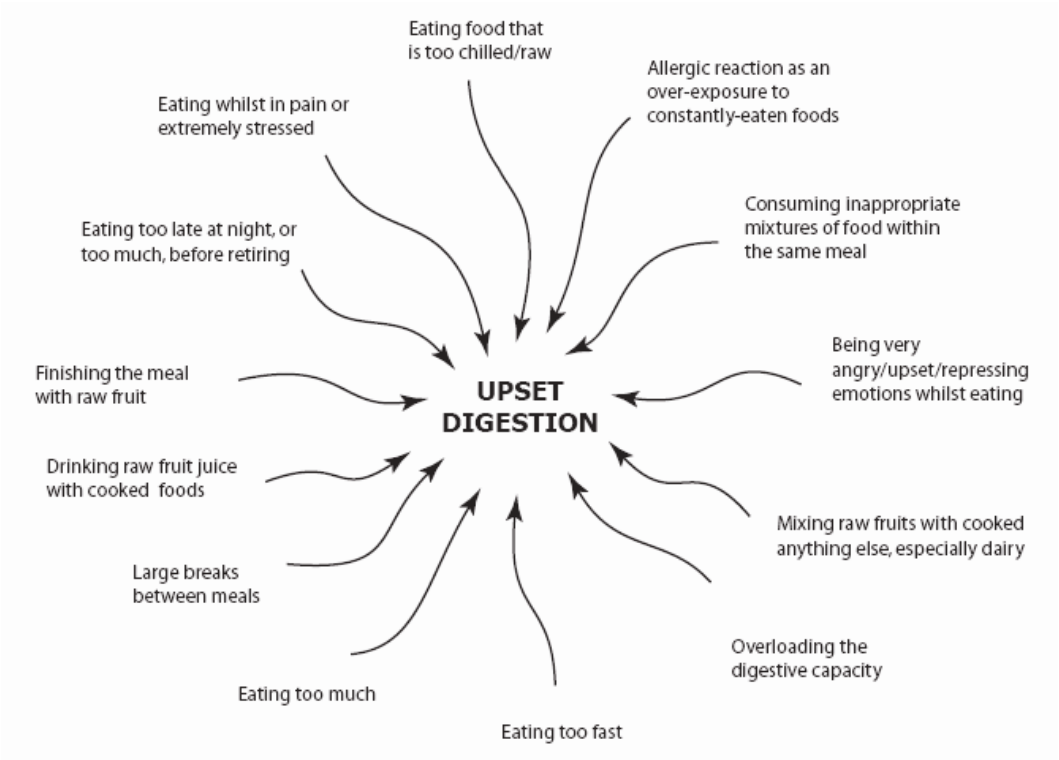


Phlegm is a more serious matter - and behind a great number of health problems - that take decades to arrive – but can be seen starting up and travelling down this road is not a permanent or a necessary route – **you can back out of it.**

It affects all of who you are – especially the choices to stay stuck/procrastinate through your life.



The most important 'cure' here is to stop making the phlegm. Firstly – what is being eaten? Temperature of what goes in your mouth, the frequency (not great times between) and moderation in all things plays a huge part here. Then – mixing up foods that just don't mix here. I am also using the naturopathic model.



Please be aware that though you may have done some of these forever, they may well be part of why you are not feeling on top of the world - and maybe never have.

Can you recognise ANY of the habits from the diagram above? If so, stop immediately - your body needs all the help it can get.

Seeing these apparently normal things that you have done all your life in a different light may trigger you to stop unknowingly making things worse – when they could be better because you have done all sorts of things – not knowing that, for example - cold makes the Spleen Qi (nausea, digestion weakening, allergies - **hence poor quality sperm**) even weaker.

Drinking anything cold, or being in a draft is enough to weaken your digestive energy and start the process going again. Help is at hand by changing your habits. Nutrition is the key – after you are calmer and the gut is working as it is supposed to.

Alcohol and self medicating with substances is not the way to help yourself become healthier – or make healthier babies.

Not orally – but using magnesium oil. Get some to absorb through the skin – it works better. The importance of magnesium to humans.

It is irrelevant if it is how you always have lived – **better sperm through life change.**

REMINDER - Make sure you are poohing at least once daily - if not – take more Vit C as bowel tolerance is actually what is need to flush out al the chemicals that are impacting in sperm quality. If you take too much magnesium – it has the same effect (automatically empty the bowels) - partially why I suggest topical magnesium, and Vit C sipped frequently all day - both are important for the toxin release. (Both needed to help make Glutathione also). If you can't tolerate oral magnesium supplementation it only means this – not that you don't need it – any more than if you crave sweets you must need them. Again – use the magnesium on the skin.

You should be using at least 10 grams of Vit C (go back to Chapter 1 and see just what it does) daily – a little often sipped as an additive to your water - and then see how much more vigour those little swimmers have!!

Perhaps juicing your veggies will allow these concentrated nutritional packages to assist you. Food HAS to be a large part of your answer.

Acupuncture MAY help. Please do not go to an allied medically trained professional who has tacked on this speciality as though it were a band-aid. Understanding the entire model is crucial. Just as it takes years to become a doctor of medicine, becoming an energy understanding acupuncturist is not a quick process.

The sperm generating cells age – far more vulnerable than 'aging' woman's eggs.

Time can heal why the sperm have been less than perfect - if you take charge and change what you have been doing. All the nutritional changes alluded to in the first chapter need a sensible life and a gut that CAN extract what it needs from what you choose to eat. There are many great products that assist pulling out the contaminants in a quicker fashion – Glutathione is the cheapest way and fastest in my clinical observations.

A very effective Glutathione accelerator (you want results soon) It is sourced through this site and can be an amazing change in all your lives - especially if you take the

entire package: their multi and a gut/immunological enhancer daily.) Under $200 for 3 months worth of all three sent three monthly to you)

Beyond this, there is a lot that your partner can do to help.

The allied eBooks – *Wake Up Your Legs* and *Releasing Natural Flows* are equally important. Some of the massage and moxa moves are to be found here.

These basic easy home-based interventions can make all the difference instantly.

Ideally the moxa is sourced through an acupuncturist – you want the smoky stick, not a 'smokeless' variety – the results are to be had with the real deal. The relief and soothing warmth to be had is not found with any other venture. The stick is put out by smothering with aluminium foil, and lasts for months.

6 – FOOD AND SUPPLEMENTS

This is a mud-map and is not intended for more than interest in order to help you put together how the entire show works. Without strong digestion and a happy life, you may find that regardless of what goes in – the outcome may not be as great as you would like. The factors allowing you to be well and healthy are covered elsewhere. Live Well package. How you can maximise your pelvic circulation is also covered in detail here.

As a man who is looking to improve his contribution to his future children, of course if SHE were also following this programme the pregnancy she has and the children you make together will be far less needing medical assistance – throughout their lives.

Please review the chapters on how a well gut works.

No gut is going to be able to work well if there is substantial stress. The stress leads usually to not so good sleep, meaning regeneration and rebuilding is less than optimal, and accelerated aging takes over. Starting here and getting great sleep – where you wake up refreshed – is vital.

Protein - Make the body and regenerate with this.

Divided into 'primary' and 'secondary' in the past.

Primary means all of the 8 amino acids we cannot make ourselves are there on hand. These come form anything that comes from a creature that drew breath. Does not have to be their flesh – can be their products. This means that as a vegan (no animal products at all) when making babies, there is a special interest in making sure there is sufficient for you before you are to make what will be the foundation for another.

It is possible and you need to be crucially aware of protein combining - and in the same meal and fat intake. Also the strength of your gut – possibly look into fermented foods as a way of life.

Personally I suspect the Weston Price style of eating especially laid out in the new book Nourishing Our Children by Sally Falloon.

I agree with author Sally Falloons' position of the NOT taking of supplements - yet who is living a life that supports life – and great nutrition in pregnancy – so the ideal is growing your own and living in a self sustainable community where food is sourced from less than 100 km away and bought where necessary in growers markets – and cooked from scratch at home - and most do not live this way.

The second best way is to ensure that the gut is working well – enzymes from fermented foods **first** after the gut has been 'spring cleaned' through removing all parasites, and mould/fungi that take hold whenever an anti (life) biotic is taken. **After this comes** – choosing food grown in the soil that is well. Are the nutrients even in the soil – let alone the plant? **Thirdly** - Can you assimilate what you have chosen to eat after breaking the food down to its constituent parts? Good questions all.

Answers:

1) – See a naturopath or natural health care practitioner who has a programme to ensure that your gut is pristine. This is a must for all – not just those who are seeking fertility care.
2) – Check out sources of biodynamically grown and with love, not artificial fertilisers on gigantic industrial farms.
3) – Fermented foods, good food combining principles.
4) - A great array of those minerals in you already – Zinc, Magnesium, Iodine and the like – to ensure that you can actually digest foods at all.
5) -The right temperature to work the food factory – the chapter on digestion is in here for you to absorb this information – it Is not available in any other works as all look to the end results – not the programmes that are biologically installed within us to have all work automatically. As we no longer listen naturally to the body, sometimes all that is needed is to go back to basics – and without the gut working well, regardless of the most perfect foods and supplements and intentions - nothing as you would imagine will come of it all as you are not using the body as per the 'owner's manual' – that you were not given out with that body.

Please stop here and read the chapter BEFORE if you have not already

There is no substitute for simple and natural living. Our forebears had far too many children, often far to close together – life was not as toxic and although hard, all were likely to net a baby with regular sexual contact. Here are some of the ingredients in food – and why a very plant based diet in a simple life helps. Fat is essential for life and most fat soluble vitamins are void without his and sunlight – or at least a large dose of Vit D is supplement form – 1,000 iu per day is nowhere near enough.

VIT D

All vitamins and minerals are interdependent and many are deficient in a 'well' body. Can't live without it. Many paths to investigate here and here for you.

Needed for the absorption of all the fat soluble vitamins (everything that is not B or C) and all magnesium. Without Vit D the entire hormonal basis for life is interrupted. Lack of exposure to sun will cause sun cancers.

Water soluble vitamins

Vit B and C are the only ones that are water soluble, meaning a little often is needed as they are not stored within any body tissues and are in constant use – thus constantly needed.

VIT B

All are needed to use any appropriately. Most studies look to the Folic Acid – (B9) and B12 as being the major ingredients - yet all must be consumed to get any benefits. The entire Vit B group are needed in the body for repair and particularly for nervous regeneration.

ANTIOXIDANTS

Needed to help stop the inner rusting as oxygen, whilst needed for life is also a very strong aging agent.

VIT C

Ever since Albert Szent-Györgyi, MD, PhD, first isolated ascorbic acid and identified it as "vitamin" C in the late 1920s, controversy has ensued. By definition, a vitamin is categorized as a micronutrient, an essential element that human bodies need in small quantities. But Szent-Györgyi made his classification before the sweeping notion that all vitamins are micronutrients took hold, and his evolving suspicion that ascorbic acid is needed in much larger doses has been shared and strengthened by many other noteworthy scientists who followed, as Hickey and Saul so ably illustrate.

Here is Irving Stone, PhD, who determined that ascorbic acid was not "a vitamin at all, but an essential dietary factor" and first proposed that high doses be given at short intervals. And Frederick Klenner, MD, known for his remarkable use of treating polio successfully with megadoses of vitamin C during a 1948 epidemic. Lendon Smith, MD, risked his reputation when he prescribed megavitamins, including vitamin C, for children in his groundbreaking 1979 book *Feed Your Kids Right*.

At the forefront, of course, was two-time Nobel winner Linus Pauling, PhD, whose popularization of the need for high-dose vitamin therapy opened the door to nutritional therapy and lifesaving "orthomolecular medicine." The list of acclaimed scientists and physicians who have championed the role of vitamin C in fighting disease is long – and includes famed orthomolecular pioneer Abram Hoffer, MD, PhD, who provides a foreword for the book – and yet, despite the gains and conclusions reached by these august researchers, **the use of vitamin C remains continually fraught with contentious resistance from the medical establishment**, which can’t quite let go of the "micronutrient" definition.

So, in *Vitamin C: The Real Story*, the authors revisit the definition of vitamin C. They make the case that **vitamin C is not a micronutrient needed in trace amounts administered once a day to stave off diet-induced deficiencies. Instead, they argue, it is a vital nutrient that cannot be assimilated from even the best diet in the significantly large amounts needed.** When the quantity of vitamin C consumed is too low, the result is illness. Scurvy is perhaps the best-known resultant disease, but the authors argue throughout this book that "almost every chronic disease has been related to an insufficient intake of vitamin C."

The real meat of this book lies in Chapter 3, "Taking Vitamin C." Here **the authors address the central debate regarding vitamin C usage: the optimal intake**. They look at how and why the Recommended Dietary Allowance (RDA) – also referred to here as the "ridiculous dietary allowance" – was wrongly determined, setting up a seemingly endless battle between those who believe that once an RDA is established, it’s a "proven," irrevocable fact, and those who have witnessed the extraordinary results effected by much higher doses. Most instructively, **the authors make their case by explaining the ins and outs of vitamin C absorption**

(demonstrating the need not just for high doses but also divided dosages), detailing the forms of vitamin C, and debunking warnings about potential side effects.

Look further into the role of Vit C and life through browsing through the massive information here - http://www.doctoryourself.com/klennerbio.html and here http://www.whale.to/vaccines/kalokerinos.html. The eBook instantly downloadable freely http://www.vitamincfoundation.org/stone/ by Dr Irwin Stone shows you just how much nature is being ignored, and how easy health and the freedom of disease really can be.

Basically Vit C is used as when stressed - the more stress - the more Vit C is needed – constantly. It is not stored in the body and humans are one of the few living organisms that cannot make it themselves. Without Vit C the liver cannot detoxify all the wastes and chemicals that are present. The collagen repair needed for life falls over and the body is subject to myriad attacks from all virus, bacteria and the like. Vit C in at least amounts of gradated 10 grams over a day – every day - are needed for optimal health – and when stressed or ill, substantially more may be utilised. The lack of this alone may cause most problems we experience.

As you will see by reading some of the research (always seemingly done with only one factor – as though food is only containing one ingredient), the application of this alone can almost instantly result in better sperm outcomes.

VIT E

Crucial as all vitamins are for life. As a fat soluble vitamin this will require a decent amount if Vit D to be absorbed to be used. Most see a difference in their overall health if they start taking this as a daily supplement – preferably with fat in the diet. Found in foods that most shun. Vit E and Vit K (and magnesium) are plentiful in dark green leafy veggies - eating far more colour in your plant based diet will straighten out most problems.

GLUTATHIONE

The master anti oxidant in the body, that must be continually synthesized. It cannot be taken orally except as a Glutathione accelerator. If you choose to take this orally, almost instantly you are likely to feel like a new person. It is about the most amazing supplement I have ever encountered – you take 6 capsules in the morning and the body continues to recycle it all day. See here for more.

VIT K

Found in all dark green leafy veggies, and needing the person to have a gut that is working with proper biological assistants (we have to have myriad bacterial helpers there) Vit K is necessary for arterial and vein health. (The 'K' stands for the German word for coagulation – can't without it). Any bleeding problem – especially menstrual and particularly if the woman has been on antibiotics for a while – the over bleeding may be caused through the imbalances by killing off all the needed good bacteria. Bones need Vit D, magnesium and Vit K all more than the calcium all are told to take.

MINERALS - MAGNESIUM

I have numerous links all through this work – please start here.

Micro nutrients

SELENIUM

Selenium is a trace mineral also invaluable for detoxing in the body, as it is needed to make Glutathione. This means it is an anti aging and anti infective agent. It is best taken in supplementation form with Vit A or E. It is needed by your thyroid to help convert the T4 to T3. Without this, your metabolism will be sluggish, allowing less likely great gut function, which in turn means poor assimilation of all nutrients. Without Selenium, a body is far more likely to suffer from most cancers – little was known about its essential presence until recently and often it is still called a toxic substance. Very little is needed and often soils are deficient in it (as in Australia and New Zealand) necessitating its supplementation for human and animal health.

ZINC

Without Zinc, (as Selenium, a trace mineral and deficient in soils in Australia and New Zealand so very often highly needed as a supplement), life is not possible. As Selenium, Zinc is also needed to assist in the conversion of T4 to T3 in the body so the metabolism works properly. Men need Zinc to assist make sperm. It is also highly needed to assist all catalytic enzymatic activities in digestion, is used in wound healing, in growing hair, assuring skin is clear (part of the gut/skin reactivity complex) and the immune system working well.

IODINE

Without sufficient the thyroid hence metabolism and digestion are incapable of functioning. Iodine is the basis for all thyroid hormones. This then cycles back to the BBT taking - if your rectal temperature is less than 36.6/8 C then it is very possible your thyroid is not supporting your gut, immune system or general health. Getting it to raise through a lot of exercise, and eating no grains, but heaps of veggies, fat and protein, as I explain in the Supercharge Your Sperm eBook will assist both your metabolism and your fertility.

7 – GLOSSARY

Please also go to the banners on www.HeatherSays.com a more comprehensive glossary/explanation is found here.

Qi - pronounced "chee", loosely translates as 'energy' but is far more than what we in the English language or conceptual fields consider to be 'energy'. In Chinese medicine, the body is seen to have many different types and uses of and for Qi. Our personal Qi circulates within and around us. It can be sort of seen as being our power source.

Blood – More than the red liquid that runs if we cut ourselves, Blood is the concept of what nourishes and renews life. Its energy allows sanity, sleeping, memory, thinking and all expressions of self to be coherent and make up who we are seen to be. Without good quality Blood the Shen has no home. Blood chapter See more.

Shen - Our Shen, whilst said to reside in the heart, is protected through the pericardium energy. This is how we are and who we are seen by others to be – all our behaviours and projections come from here. is a Chinese concept that covers the essence of self in a spiritual sense. Shen governs who we are and how we express ourselves. A well-nourished Shen allows us to present a consistent personality to the outside world. Our Shen allows our clarity of speech, our brightness of eye, our coherence of thought, and our ability to live in mental and emotional tranquillity.

Jing - Jing is the essential energy building block. It forms the foundation of the Kidney energy, which is itself the root of Yin and Yang Qi (energy) above. Like any another form of inheritance, Jing can easily be spent unwisely, when not adhering to sensible guidelines.

Yin - all aspects of life can be polarised into Yin and Yang. The yin portion of anything is more solid/substantial and dense. In the body this translates usually as 'Yin Qi' meaning that which is cooling, moistening, allows rest and regeneration and that which is more nutritive - blood and body fluids being examples.

Yang – as above. The more yang component of anything is that which has less form, less structure and is more ethereal/resonant.

Yang Qi is like the metabolism master switch – also determines the strength of the immune response, heating, digestion and circulation.

Liver Qi – Liver energy must flow freely; otherwise all aspects of self are affected. Similarly, all expressions of self and blockages therein affect how the Liver Qi can continue with keeping all flowing as designed.

Spleen Yang Qi - Our Spleen Yang is that which controls our food assimilation and hence the quality of all our Blood and other raw ingredients. The Spleen Qi also holds all our tissues, organs and Blood in place. When upset – especially by the invasion of cold, or by excessive 'stress' - when our Liver Qi gets too upset goes horizontal and attacks the Spleen directly – this function, as with all the digestive

ones, and our abdominal contentment can just give way. This may lead to hernias, varicosities, prolapses, bleeding problems, and gut disorders – heartburn, reflux and IBS being the common ones.

Our lymphatic and immune systems are totally dependent on our Spleen Yang. All cancer and other illness issues are directly tied to the health or otherwise of our gut. When the Spleen energy is weak, we start getting diarrhoea, maybe always loose stools, and if more severe – with indigested bits in them.

Kidney Qi - The Kidney Qi is that which all other aspects of the body depend on to function. The root of our being is the Kidneys and only part of their job is urine output – more the task of the Lung and Spleen energy – hence when an acupuncturist speaks of your Kidney energy, think of your hormonal and neurological wellbeing.

Kidney Yin – that aspect of the Kidney energy that allows regeneration and life. It is in balance with the Kidney Yang below. When people are aging too fast, when they are exhausting themselves with chemicals and substances, fast living and not sleeping, drinking enough water - instead wasting their inner resources, it is the Kidney Yin that keeps all moist and vibrant that is used up. This leads to hot feet at night, to women being told that their egg quality is low, and for men – not good sperm quality –all is remediable – with time and all the supplements mentioned in Chapter1.

Kidney Yang – that which gives the sexual oomph and the vigour of the sperm. It is also needed to feel desire, to be sexually motivated and capable, and to be able to orgasm.

Wei Qi - a particular type of Yang Qi protects us. This is called Wei Qi (energy) and works on many different levels. Wei Qi circulates through the muscle and superficial layers of the body and energy fields, in a constant tidal flow. It has to retire internally at rest, to be replenished as part of the overall sleep /recharging cycle.

Meridian – energy travels through, along and within the body in lines or meridians. These form a grid often seen in acupuncture clinics as maps of the body.

Pericardium - Our **pericardium energy function** begins at the first breath we take independently at birth. At this moment our previous lifeline from our mother shuts off, and we begin supporting ourselves. This instant, and the ease or shock that it brings, sets in motion who we become. The imprint of the effort or grace at that moment reflects upon us from then on and affects all the perceptions we ever feel through our senses throughout life.

Damp – the energy of obstruction that seeps into all aspects of self – be it cloudy head, muddled thinking, fat and cellulite that settle in and sit there regardless of your best efforts to move it, phlegmy throat, discharges. Candida, inability to get out of the space you are in . .

Phlegm – as a worsening of the presence of damp. Phlegm is more solid – and obstructs even better than the damp. Insubstantial (No form) phlegm is what clouds the senses and has people seem ‘mad’ or ‘out of it’. The more solid phlegm is that which you can cough out of the lungs, or see as discharges, pus and cholesterol in arteries.