Acupuncture explained in terms of hypothesized biochemical mechanisms with copper-, zinc and magnesium ions.

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(Dutch version on website includes ATP generation and cellular production of heat. This is not includes in the pdf.)

ABSTRACT

In this paper a biochemical hypothesis is proposed for the working of acupuncture. In this hypothesis, copper and zinc ions play an important role. Inserting a needle into an acupuncture point will locally damage the tissue. It is known that this causes a local release of zinc ions. It is also known that both zinc ions and copper ions bind on the intracellular transport protein metallothionein, where they compete with each other. Furthermore, the copper dependent enzyme lysyl oxidase plays a role in the change of softness and stiffness of connective tissue. Needling into an acupuncture point can change the structure of connective tissue. Locally soft tissue becomes stronger, and locally stiff tissue becomes softer. From this is hypothesized that needling an acupuncture point can affect its tissue structure by biochemical reactions that involve copper and zinc. Subsequently it is hypothesized that the proprioceptive system projects the pattern of soft and stiff places of all acupuncture points onto a neural map in the brain, on which meridians are formed by associations of subgroups of acupuncture points. A meridian is interpreted as the mapping of a particular set of skin locations above muscle-groups which is associated with a certain behavior. Examples are the stomach meridian, related to eating; the gallbladder meridian, related to performing physical work or to capturing a prey; and bladder meridian which relates to changes between lying and standing. Then the activity pattern of the neural map is associated with a particular physiological state and sensory patterns, and the brain responds to the activity of this neural map with a regulation of the autonomous nervous system (vagal nerve) and a regulation of neurotransmitters and hormones.

We hypothesize that the local stiffness of acupuncture points indicate certain physiological states of the body, related to copper or zinc. In particular zinc dependent processes like parts of digestion, and copper dependent processes like production and degradation of dopamine. Because during the day, different physiological processes are planned, both copper and zinc vary during the day. In this circadian rhythm of copper and zinc, the entero-hepatic circulation and the cerebrospinal fluid is involved. The copper and zinc displacements due to these regulations are then in turn hypothesized to affect the structure of tissue around related acupuncture points. Concepts in acupuncture are related to the copper and zinc hypotheses: the Wu Xing model, yang, yin and blood emptiness, backshu points, shu transporting points, Qi stagnation, damp, phlegm, external pathogen factor and Jing. Adding a relation between copper and iron ions leads to an interpretation of yin emptiness and blood emptiness. Adding the fact that magnesium competes with zinc leads to an interpretation of Wei Qi and Front Mu points. Magnesium is important in phosphorylation of diverse enzymes, a kind of energy potentiation of enzymes, and in binding NADH and NADPH as cofactors. This may be associated with the term "Yin" used in Traditional Chinese Medicine. Symptoms of magnesium deficiency are described in symptoms used in acupuncture diagnosis. In summary, both magnesium and copper may
show yin characteristic, copper with heat, stagnation and restlessness aspects. Finally, effects of copper, zinc and/or magnesium are associated with the “Three Burners: Upper Burner, Middle Burner and Lower Burner” respectively.

1 INTRODUCTION

Acupuncture and Traditional Chinese Medicine (TCM) on the one hand and conventional Western medicine on the other hand have completely different views about the functioning of the human body. Western medicine is based on conceptually fundamental and verified models from the exact sciences such as physics, chemistry, and biology. TCM is also based on many experiences and observations, but its theoretical fundamentals are completely different and unrelated to the Western fundamental sciences. However, since both medicines have the same human body as subject, it should be possible using biological, biochemical and physiological mechanisms to understand acupuncture in a Western way. This paper proposes a biochemical hypothesis to understand the working mechanism of acupuncture in Western physiological and biochemical terms. Herein, copper and zinc will play an important role. This article is intended to stimulate research on the effect of acupuncture and is intended to be a small step in theory of acupuncture. Which diseases and circumstances are suitable for acupuncture? Under which conditions does acupuncture contribute to a regular treatment? Chinese patterns of acupuncture and Western medicine patterns differ in their concepts. Acupuncture works with concepts such as for instance meridians, heat, cold, fullness and emptiness, damp, and egm [1]. The choice of acupuncture points on different locations, called the needle formula, is determined by diagnosis, derived from local aspects (related to the complaint) and global aspects, characteristics and symptoms of the patient. A diagnosis first will rely on a questionnaire. Questions in acupuncture are for instance about appetite, and the question if the client show a quiet or restless behavior. The color of skin and tongue characteristics will also contribute to the diagnosis used by acupuncture. The acquired diagnosis gives an indication of the needle formula: which acupuncture points should be needled. Furthermore, when tissue around an acupuncture point is too loose or too hard (measured by palpitation), this point is a candidate for the acupuncture formula, at least when this point is related to the diagnosis. This paper is organized as follows. Chapter 2 motivates why copper and in particular zinc are central factors in the hypothesis of this paper. Chapter 3 discusses a number of local effects of needling. It describes hypotheses of how needling an acupuncture point can affect its tissue structure by biochemical reactions that involve copper and zinc. Chapter 4 describes how and where stiffness and softness patterns of acupuncture points are mapped in the brain. Patterns of acupuncture points are related with the five types of behavior as is known in acupuncture. These patterns are interpreted as patterns of muscle activity in the context of certain behavioral modes. Chapter 4 also addresses the concept of meridians in acupuncture. Then in chapter 5, biochemical functions of copper and zinc are related to physiology. Influences of certain disorders and drug (side)effects on copper on zinc are describes. In chapter 6, the Wu Xing model, used in acupuncture will be described in the context of the copper and zinc related biochemistry. Furthermore, chapter 6 describes emptiness in yang, yin and blood expressed in copper and zinc terms. Chapter 7 ends with discussion and conclusion.
WHY COPPER AND ZINC AS CENTRAL FACTORS IN THE HYPOTHESIS?

Central in the hypothetical understanding of the working of acupuncture in biochemical terms are zinc and copper ions. Why? Yan [2] measured around an acupuncture point an increase of zinc and copper ions. Yan measured in three of the four acupuncture points showed significantly elevated concentrations of elements Ca, Fe, Cu and Zn in relation to levels in the surrounding tissue, with similar elevation ratios for Cu and Fe. In this article we first will concentrate on copper and zinc ions. Chapter 6.1 includes an effect of calcium in the context of copper and zinc. Chapter 6.2 includes iron in the context of copper, and Chapter 7 includes magnesium in the context of zinc. Zinc ions compete with copper ions on their binding protein, metallothionein [3]. There are a number of copper and zinc dependent enzymes. This article describes how some of these enzymes could be associated with symptoms that play a central role for diagnosis in acupuncture. Why are copper- and zinc ions such presumed important cofactors? Recent research of copper and zinc reveal interesting results about contribution of a role of copper and zinc ions in nerve conduit [4], and nerve condition during inflammation [7], a role of copper in cell signaling pathways [5, 6] and a role of zinc in DNA expression. Copper and zinc are accepted as signaling molecules and not just "trace elements" [5]. Frausto da Silva [8] postulated that copper and zinc ions are incorporated in organisms at different times during evolution: zinc at the time that organisms became multicellular, and copper at the time that motion developed. Frausto da Silva’s story interpreted in the context of our article, goes roughly as follows. During evolution oxygen increases in the environment. First, this leaded to a biological role of zinc, and secondly, later copper received a biological role. With respect to zinc: due to the greater amount of oxygen in the environment, zinc sulfide oxidized to free zinc ions and sulfate. As a result, free - zinc ions appeared. The development of multicellular organisms with a digestive tract was created possibly as follows. Zinc was used as a cofactor in the hormones that controlled digestion. Each cell was supplied with sufficient glucose: insulin that affects the glucose input into the cell is zinc dependent. With a meal, also badly digested or wrong parts pass to the intestines. The immune system therefore had to act after eating. Zinc ions as cofactor play a major role in the immune system as well as the digestive system. With respect to copper: copper sulfide is more stable than zinc: the electro negativity in Pauling scale of zinc is 1.65, of copper is 1.90. Therefore, first zinc was released as result of increase in oxygen. But with the further increase of amount of oxygen during thousands or millions years, the copper sulfides in the environment oxidized finally to copper ions and sulfate. As a result, free copper ions appeared. For intensive movement, speed, power and timing, needed to catch a prey much energy is required. The molecule ATP (adenosine triphosphate) is the ultimate, readily available energy source of many biochemical reactions. ATP is made, inter alia, from glucose. The second last step of this ATP synthesis (complex IV) is catalyzed by an enzyme with a copper ion as a cofactor (cytochrome C oxidase). The rate of ATP production increases by an increase in copper ions. Research demonstrates that a brain copper deficiency results in mitochondrial dysfunction mediated by a decrease in the activity of complex IV [9]. Possibly a slight variation in cofactor concentration is also a healthy physiological regulation. In addition the third last step (complex III) of ATP production is inhibited by free zinc ions [10]. Thus, in ATP production, zinc and copper exhibit an opposite effect. In summary, Frausto da Silva postulated that during evolution, zinc appeared as a cofactor during the transition into multicellular organisms with a digestive system and an immune system. Copper may have appeared as cofactor during the transition to moving organisms. Of course this is a rough sketch. Because other organisms like plants and bacteria continually changes during evolution and also includes copper in their mitochondria for instance. Cofactor are very important, they are very
reactive, and influences the structure of proteins. When an enzyme has enough cofactor, for instance copper or zinc, it works most efficient. Besides, zinc and probably also copper influences expression of zinc (and copper) dependent enzymes. Central in this article is a different need for copper and zinc during the day: it argues for a circadian rhythm in copper and zinc ions, with for each a different separate maximum and a minimum during the day. Some flow, transport in copper and zinc is needed, to let enzymes who need these ions as cofactor work most efficient. This paper describes how acupuncture in the needled location biochemically should perform his effect, by manipulating zinc in first instance.

In this article we describe "zinc", and "copper", but in detail, the body is more complicated. Mentioned research in this article is sometimes based on serum zinc concentration [1b]. Serum zinc is relatively constant, except during long-lasting periods of dietary zinc deficiency, long lasting severe stress response, such as in cases of cardiac infarction, infection or inflammation, or chronic emotional stress, which cause the serum zinc to decrease [1b]. To determine if deficiency in zinc is present, serum zinc and lymphocyte zinc should be measured [1b]. It is important that symptoms of zinc deficiency are visible, and are in remission after supplementation of zinc [1b].

Practically 98% of the zinc is located in intracellular space [1b]. In contrast, the concentration of free zinc inside the cell is lower than in extracellular space (and blood serum) [1b]. In fact healthiness is determined by a proper membrane function [1b]. Influx in the cell of the free zinc is a passive process due to the electrochemical gradient [1b]. Transport of zinc between organelles and cytoplasm inside the cell, and efflux of zinc is (sometimes also passive but mainly) an active process [1b]. Intracellular a potassium-zinc anti-transport (and chloride-bicarbonate anti-transport) takes place [1b]. The efflux includes a calcium zinc anti-transport channel [1b].

One should be cautious with supplementation of zinc, because high intracellular zinc promote apoptosis. First it should be demonstrated that a zinc deficiency has occurred. A proper membrane function is important, and for instance potassium is also important. Furthermore, different ions interact on each other. For instance, high serum zinc is accompanied by deficiencies of calcium, copper, iron and by anemia. Hopefully, this article gives the impression that ions are important and different balances between ions act in accompany, guiding or following important physiological processes. As will be described, acupuncture may temporarily relocate zinc, to stimulate certain physiological processes.

### 3 LOCAL EFFECTS OF NEEDLING AROUND AN ACUPUNCTURE POINT

#### 3.1 Release of zinc ions.

During needling of an acupuncture point, zinc ions are released. The zinc ions do not originate from the needle, but are derived from neighborhood tissue. A number of articles suggest an increased concentration of zinc ions [2, 11, 12, 13]. Around the acupuncture point, the local painkiller adenosine [14] and nitric oxide (NO) are released [15]. Both NO and adenosine promote the release of zinc ions: NO makes zinc ions come free from MT (metallothionein) [17]. Another molecule that is measured in the serum after electrical acupuncture is the antioxidant glutathione [18], which also releases zinc ions from MT [19]. Adenosine, NO and glutathione play a role in the small inflammatory response in the little wound, generated by the needle. The released zinc ions in an acupuncture point have an effect in efficiency of certain enzymes: the enzymes with zinc as cofactor.

Metallothionein (MT) is an intracellular protein that transports copper and zinc. A known characteristic of zinc on MT is that zinc ions stimulate the DNA expression of MT [13, 22, 23, 24, 25]. We hypothesize that this also happens in the needled acupuncture point: the
released zinc ions may promote locally the expression of MT. Then, this local increase of MT can collect an increase of copper. The copper may be derived from the blood, where ceruloplasmin transports copper ions.

3.1.1. Macrophage copper and zinc

A possible role of this local increase in copper and zinc may be to optimize the functions of certain immune cells, the macrophages. The little trauma wound caused by needling the acupuncture point result in the first line of defense of the immune system, the innate (nonspecific) immune system. The injured cells produce chemical factors, like cytokines, which recruit immune cells to the site of infection. One of the immune cells of the first line of defense, the macrophages, are large eaters by phagocytosis, and (are in some places already present in the tissue), or move outside the vascular system entering the injured area between cells. These macrophages play an important central role in the immune system. For instance, a macrophage digests a microbe and presents the microbe’s antigen on its surface to alert other white blood cells to the presence of the invading particle. Other white blood cells then multiply and amount a specific immune response against this pathogen. This is done in a way that the pathogen is remembered by the immune system. Copper and zinc are directly linked to macrophage antimicrobial pathways [25b]. Enough copper and zinc in the surround of macrophages is an advantage.

Furthermore, zinc is important in platelet aggregation, coagulation, anticoagulation (combined with calcium) and fibrinolysis [25d]. The concentration of free zinc increases in the vicinity of activated platelets [25d]. Once activated, platelets secrete zinc, which provides a mechanism for propagation of coagulation [25d]. Zinc increases the fiber thickness of fibrin clots [25d].

Finally, erythrocytes, lymphocytes and neutrophils can also release zinc into the blood [25d].

Two types of macrophages, representing two stages of inflammation and cell division.

In muscle injury, macrophages remove debris and promote the proliferation and differentiation of parenchymal cells [26b]. M1 macrophages release inflammatory cytokines, mediate defense of the host from bacteria, protozoa and viruses, and have roles in antitumor immunity [26b]. Alternatively activated macrophages (M2) have an anti-inflammatory function and can secrete interleukin-10 [26b]. Onset of inflammation primarily through the M1 phenotype and then can switch to an M2 phenotype to promote healing and repair [26b]. Acupuncture has analgesic and anti-inflammatory effects. [26b] Acupuncture in a mouse model of peritonitis increased circulation levels of IL-10 [26b]. The treatment of acupuncture consists of repeated daily treatment of SP-6. Prior data show that administration of IL-10 inhibits the development of hyperalgesia in several animal models of pain [26b]. Immune cells attracted to the site of inflammation mediate the change in phenotype by acupuncture [26b]. Macrophages are plastic and can differentiate into different subtypes including M1 and M2 [26b].

Both a diet with zinc deficiency as well as a diet with copper deficiency result in an impaired immune system, which effect is reversed when zinc or copper respectively is added in the diet [25b]. We hypothesize that first a copper stage takes place, which is followed by a zinc stage. During the copper stage type M1 macrophages are active, during
zinc stage, type M2 macrophages are active (which is described in next alineas). Type M1 attacks pathogens by NO and phagocytosis of debris (of the inflammation) is performed. During inflammation: edema and pain is present. Type M2 stimulates cell division and cell growth. A transition from M1 to M2 may reduce pain caused by edema.

The first stage, copper stage is probably initiated by the liver, responding to cytokines send by damaged cells in the needled acupuncture point. The liver responds by increasing synthesis of ceruloplasmine (a copper transporter in serum) and decrease synthesis albumin (a zinc transporter in serum) [73b]. This may be locally felt at the needled acupuncture point. In the first stage, TNFalpha (secreted by M1 macrophage) promotes the accumulation of copper within the phagosomes [25b]. This metal may have some role in macrophage antimicrobial responses [25b]. Copper may regulate both immediate and delayed macrophage antimicrobial pathways [25b]. Copper may be used during delivery of copper to target pathogens for destruction [25b]. The copper may be used (in combination with ceruloplasmin, a copper binding protein) for iron export from macrophages, as an additional defense mechanism by restricting bacterial growth [25b].

However, zinc still plays a role in the “copper phase”: zinc appears to enhance the microbial activity of macrophages. During this so-called copper stage, serum zinc shows a lower concentration, but intracellular zinc is still present and very important, zinc is needed to enhance the microbial activity of macrophages [25b]. The focus to zinc signaling in monocytes and macrophages has been on Toll like receptor signaling: downstream of this: zinc regulates inflammatory gene expression through multiple pathways, including tyrosine phosphorylation, MAPKs (mitogen-activated protein kinases), protein kinase C, phosphodiesterases, and NF-kB [25b].

In the second stage: zinc is limiting excessive inflammatory responses, which may be deleterious to the host [25b]. How increases zinc? The concentration of free zinc increases in the vicinity of activated platelets [25d]. Once activated, platelets secrete zinc, which provides a mechanism for propagation of coagulation [25d]. Zinc increases the fiber thickness of fibrin clots [25d]. We hypothesize that these platelets are responsible for the transition to the second stage, the zinc stage. This zinc promotes the coagulation of blood platelets. Possibly a certain glycoprotein present in blood is involved in this process, histidine-rich glycoprotein (HRG) [26c]. Heparin is an anticoagulant. HRG may bind to this heparin, (inactivation) which promotes coagulation [26c]. The formation of HRG-heparin complex is enhanced in the presence of zinc [26c].

Furthermore, zinc upregulates ant-inflammatory interleukin (IL-10), and down-regulate proinflammatory cytokines (TNF alpha, IL-1, IL-2) [26d]. This fits in the stages: IL-10 is secreted by M2 type macrophages. TNFalpha is secreted by M1 macrophages.

Depending on circumstances a macrophage can develop into M1 type (secresing TNFalpha, among others) or in M2 type (secresing IL-10 among others). An increase in zinc may be one of the ways of regulation to change this macrophage biochemistry. From arginine, M1 produces NO. M2 on the other hand, produces from arginine ornithine.

With respect to stage 1: NO synthase is a key enzyme in NO synthesis. Zinc has an inhibitory role as a regulator of iNOS activity in inflammation [26e]. (NO synthase). Zinc down-regulates the expression of iNOS. (in endothelian cells) [26e]. While pro-inflammatory stimuli including LPS, IL1beta, TNFalpha (secreted in stage 1), induce iNOS (=cytokine mediated) expression, via activation of NF-kB [26e]. With respect to stage 2 of zinc optimum, on the other hand, zinc increases ornithine decarboxylase activity, a key enzyme in the synthesis of ornithine [26f]. Ornithine is involved in cell division.
It is shown that acupuncture (producing a very small wound, with not much to clean for M1) stimulates the transmission from a situation with a majority of M1 type macrophages to a situation with a majority of M2 type macrophages and the presence of IL-10 [26b, 26g]. In acute inflammation, like arthritis but also in atherosclerosis a majority of M1 type macrophages is present. In tumors, a majority of M2 type macrophages is present.

3.2 Zinc changes properties of the local connective tissue in the acupuncture point.
The measure of stiffness or softness can vary among individual acupuncture points, and is part of the diagnosis tool in acupuncture. These stiff locations are the result of use of muscles during a period of physical work, which makes the acupuncture point in the neighbor of these muscles stiffer. A period of inactivity makes the acupuncture point softer. By proprioception, the brain measures the stiffness of all acupuncture points, and associates this with recent behavior. This information is used in subsequent behavior, and will be associated with neurotransmitters and hormones. Acupuncture changes the local stiffness and softness of the tissue of the acupuncture points into normal values. This is performed by the copper-containing enzyme lysyl oxidase, which is related to the rigidity of the connective tissue, a measure of elasticity. The enzyme lysyl oxidase makes mutual connections (cross links) between collagens and elastines of the connective tissue. The presence of many copper ions results in hard working lysyl oxidase molecules, and in a stiff “replete” tissue. Few copper ions result in loose, very elastic and pliable, soft, “empty” tissue.

Repletion.
Repletion or stiffness in the tissue at the acupuncture point arises from many copper ions, in fact: many active lysyl oxidase enzymes. Stiffness arises after a period of physical work. During physical work, an increase in glucose and ATP is required, and thus an increase of the copper-dependent enzymes that synthesize ATP and the zinc-dependent enzymes that transport the glucose through the cell membrane. Zinc ions play a role in the availability of glucose by contributing to the efficiency of insulin receptors. Insulin promotes the uptake of glucose by the cell. Enzymes with copper ions (cytochrome C oxidase) are involved in the production of ATP. More enzymes means: many copper and zinc ions are in local circulation. When we stop with physical work, fewer enzymes with respect to glucose and ATP are needed, so fewer copper and zinc ions are needed. In which direction does this overflow of zinc and copper ions go? MT binds preferably copper ions. So, after stop of physical work, MT prefers relatively more copper. We assume that MT transports the copper to lysyl oxidase. Then an increase of copper occurs, which results in more efficient lysyl oxidase and thus in repletion. Needling will dissolve this repletion: In repletion, MT is bounded with a relative large proportion of copper ions. By needling, the released zinc ions stimulates the DNA expression of MT. As a consequence, there is enough MT to bind zinc ions also. The ratio of copper to zinc in MT will therefore go back to normal proportions. MT binds relative less copper ions, lysyloxidase becomes less active, and the tissue structure will become normal flexible. Hypothesis: the ratio of copper to zinc in MT determines the velocity of production of lysyloxidase. This determines locally the stiffness
Emptiness.
Locally soft tissue in an acupuncture point is called "empty". Emptiness arises after a chronic inactive period: muscles are less used, less ATP is needed, so less copper and zinc ions are in this local circulation. Because synthesis of MT depends on zinc ions, the acupuncture point contains less MT. The fact that this situation was chronic, implied apparently that copper is transported away to other parts of the body. Little MT (and copper) brings not enough copper to lysyloxidase, and the connective tissue obtain a loose structure. Needling will dissolve emptiness, because the released zinc increase stimulates the synthesis of MT. MT searches for copper ions (received by ceruloplasmin in the serum) and brings them to lysyloxidase. MT binds copper by preference. The hormone epinephrine assists by releasing copper ions from the liver (by releasing in the liver the transport protein ceruloplasmin, [20]). In the case of emptiness, the number of MT determines locally the softness of the connective tissue. Synthesis of MT and lysyloxidase are changed by needling. Now we can describe how strong needle technique or too many needles result in a global side effect, especially in the case of emptiness. In the case of emptiness, there is not enough zinc available. MT prefer the copper ions. This process is faster than the stimulation of synthesis of MT by the released zinc ions. The ratio copper/zinc on MT temporarily increases too much. A relative surplus of copper ions can lead to symptoms like temporarily headache, or temporarily depression [27, 28]. These symptoms may be part of side effects of strong needling.

Biochemical aspects of copper-transport in lysyl-oxidase synthesis and role of MT.
Fibroblasts are the most common cells of connective tissue. They synthesize the extracellular matrix and collagen. Fibroblasts are characterized by an abundance of copper chaperone CCS. CCS is a measure of the intracellular copper homeostasis [29]. These cells produce the copper dependent lysyloxidase. Question is, in these fibroblasts, why the ratio of copper to zinc on MT in detail, biochemically, determine the activity of total lysyloxidase, to produce the stiffness of connective tissue? MTs have been postulated to function as cytosolic copper store in normal copper metabolism ([31]). The transport of copper to lysyloxidase is describes as follow: After the syntheses of the apoenzym lysyloxidase by mRNA, the apoenzym lysyloxidase is transported to the golgi network, where its three dimensional structure is generated and the copper ion is incorporated. There are two ways that copper reaches the golgi apparatus: by import of copper ions into the cell and by recycling copper, and secondly, by a catch of the intracellular degraded copper enzymes. Intracellular, MT as a sink of copper ions, catches copper of degraded copper enzymes and delivers it to copper chaperones like Atox1 [32]. The other source of copper is extracellular. Copper is imported by the plasma membrane transporter protein Ctr1, followed by a rapidly binding to intracellular copper chaperone proteins. The copper chaperone Atox1 delivers copper to the copper transporting protein ATPase7a. ATPase7a directs copper within the trans golgi network to the proteins, among others lysyloxidase.

In fact, the total amount of MT is not only related to the number of copper ions in the cell, but also to other factors. MT binds zinc ions, and is possibly a zinc store. As will be described in chapter 6.1, this makes the intracellular MT suitable for estimation which behavior is expected at the measured time of the day. Store function of MT should be at such level that this estimation remains possible. Global processes may prevent that the
fraction of copper to zinc will be too much: too much copper (in the presence of too much bile acids causes itching, or massage. Other processes, calling for eat behavior, stress or sleep behavior may also be suitable.

In fact, the redox state of a cell or cell compartments has been shown to correlate with the redox state and abundance of Zn(II)-binding carrier metallothioneins (MTs) in cells or particular cell organelles [33]. MT and zinc are involved in a wide variety of redox reactions and Zn(II) transport in cells through reactions with other known redox active species such as glutathione and NO [33]. These events occur in response to certain biochemical triggers such as hormone release or changes in nutrient level as part of normal biological rhythms or cell cycles [33]. Zn(II) deficiency and/or elevated levels have been known to be linked with oxidative stress/ protection [33].

Question is if the fraction of copper to zinc ions on MT determines the velocity and efficiency of synthesis of lysyloxidase. Possibly the number of copper enzymes determines configuration of certain enzymes responsible for synthesis of lysyloxidase. As metal ion and cofactor it shows a possible effect. The exact mechanism how MT performs this change in stiffness is not completely solved. This important step is needed in the work hypotheses of acupuncture of this article.

### 3.3 Albumin: extracellular transport of zinc

Competition between copper and zinc mainly takes place intracellular, but can take place in extreme situations on albumin, the main transporter of zinc in blood. Albumin may also bind some copper, mainly to another binding site, but hypothetically, in extreme cases the binding of copper and zinc may show concurrence. The zinc binding site of albumin can also bind other 2+metal ions, like copper, but in principle copper has another preferred binding site [33b]. In competition experiments, there are three sites with affinity for zn and cu [33c]. Under physiological conditions, only zn will be bound to the high affinity site, as cu binds much more avidly to another (first) site [33b]. The second cu binding site in these three albumins is a multi-metal site binding for cu, ni, zn or cd, with comparable affinities, a competitive binding [33c].

Later in this article, we will describe the probably more important mechanisms to hypoalbuminemia and edema. Because zinc is mainly transported by albumin, hypoalbuminemia may decrease or slow this transport. Diverse hormones, like insulin, glucagon, thyroid hormone and growth hormone regulate albumin synthesis in the liver. This synthesis is carefully regulated, to optimize blood circulation and delivery of nutrients.
4 INTERPRETATIONS OF PATTERNS OF ACUPUNCTURE POINTS INTO FIVE BEHAVIOR TYPES

4.1 Map in the brain

Information of acupuncture points receives the brain, and is associated with five types of behavior, inspired by the classification of behavior used in acupuncture theory. By proprioception, the brain measures the stiffness of all acupuncture points. This information is projected on a neural map in the brain. There are two candidates for this map: acupuncture activates the anterior cingulate cortex and the cerebellum [34]. The cerebellum has a strong learning capacity. In the mossy fibers of the cerebellum, the highest concentration of zinc ions in the brain is measured [35]. The importance of zinc is illustrated by the fact that a severe zinc deficiency in the food yield a number of symptoms similar to the situation after a cerebellar lesion [36]. After administration of zinc in the diet, these symptoms disappear. Zinc is stored in synaptic vesicles of glutamate neurons and inhibits the glutamate receptor [37]. Copper performs its action in the opposite way. It accelerates signal transmission by inhibiting the inhibiting the neurotransmitter GABA: copper ions block the GABA receptors in the cerebellum [37b]???. These copper ions are co-released into the synaptic cleft during vesicular release. Furthermore, the healthy cerebellum plays a role in automatically adjusting the execution of emotional display to cognitive and situational context, which, if not balanced, this results in inadequate behavior. In detail it may play a role in the generation of emotionally congruent autonomic reactions [38]. So the cerebellum is involved in fine tuning in both physiological and psychological ways. Flexibility in behavior may be related to the anterior cingulate cortex. Activity in the anterior cingulate cortex is related to regulation of blood pressure, heart rate, and reward anticipation, decision making, empathy, pain and emotion [39]. Herein, dopamine is involved. The anterior cingulate cortex plays a role in reward prediction and error monitoring. The anterior cingulate cortex is more active when anticipants explore alternative options rather than exploiting a known source of reward. Modify behavior on future trials [40]. This sounds as dopamine-dependent flexibility interpreted in a later stage of our article. Finally copper as cofactor is used in peptide alpha amidating mono-oxygenase, essential for the synthesis of pituitary hormones [29]. The pituitary gland is central in hormone regulation.

4.2 Five types of behavior, five senses and corresponding meridians

On the activity pattern in the map of projection of acupuncture points in the brain, five types of behavior are distinguished by the brain. Probably also at the level of this map, meridians are constructed. A meridian is the result of an association by the brain of a specific set of acupuncture points with each other. The set of acupuncture points corresponding to a meridian mostly lies in the length direction of the body. Meridians are located around a certain pull direction of a set of muscles. Meridians represent an indication of what behavior or circumstances have been showed for a long time. They also show which behavior has been chronic absent. The five behavior types are: digestion, physical work, sleeping, withdrawal, and sociability, and will be described as follows. Digestion starts with eating and ends with defecation. When someone sees a snack, a direct move forward is made to take it. In this motion, the muscles of stomach and spleen meridian are used. The so called stomach and spleen meridians are localized at the front of the body. Muscles along this route are also used by defecation. In defecation the abdomen muscles pull together to push the stool out. This contraction also moves lymph and blood upwards. This includes the result of the digestion: the digested nutrients,
such as glucose, glycerol, fatty acids and amino acids. A second type of behavior, physical work is interpreted in terms of behavior with a goal, an orientation, for instance to catch a prey, or to run away, and to make a strategic choice. Does the prey move to the left or to the right? Mostly it is not wise to go straight to the goal, but wiser to include circumstances. This behavior, going to left and/or right is correlated with the use of muscles along the sites of the body, the location of the gallbladder meridian. A third type of behavior is a measure of alertness or relaxedness: a difference between sleep and awake, which is lying down versus standing up. Muscles that express most clearly this choice between stand up and lie down and relax are the back muscles. The bladder meridian is located around the back muscles. A fourth type of behavior is withdrawal to analyze. During withdrawal, one can form an opinion and, sort things, throwing worthless things away. This process can be physical or psychological. Thumb and forefinger are used to sort things out, and to separate right and wrong things. A withdrawal movement with the body is demonstrated by making small by pushing the arms to the chest. The chest constricts. The chest muscles contract by exhaling. By contracting the meridian of the long intestine, the arms are held against the body. The time of withdrawal and relaxedness could be a suitable time for the immune system. The fifth type of behavior is sociability. During the enjoyment of sociability, a wish to encounter people arises, and activity is focused on the outside world. When the shoulders are straightened, the body is visible to the outside world. This posture of the body makes clear for the environment what the intention of the following behavior can be. This is the muscle movement along a part of the small intestine meridian. Say hello by reaching the arms up, ready to perform a chosen behavior. Body and face are visible to other people. They can predict the next behavior. Reaching up the arms uses muscles along the other part of the small intestine meridian on the arms. The muscle movements along the Three Warmer meridian on the arms are for encountering, they are used in an embrace. They can also be used by turning to left or right to go through the outside world, and to avoid obstacles or a person, to adjust to a proper distant to subjects or persons.

These five types of behavior are according to acupuncture are not only correlated with certain meridians, but also to the functions of certain organs. The vagus nerve convey information about organs (heart, lungs, gastro intestinal tract) to the brain. This nerve measured at the level of the organs mechanical, chemical, osmotic, thermal, and possibly nociceptive stimuli: according to the copper en zinc hypotheses, the state of copper around an organ may be reflected by the stiffness of the connective tissue around the organ, determined by lysyloxidase. Sensory neurons of the vagus are located in the nodose ganglia, and glutamate is thought to be the primary neurotransmitter of the vagus nerve, projecting a map of organs into the brain [41]. An important aspect of behavior is the sensory system. In acupuncture the meridians are not only associated with five types of behavior, but also with five types of sensory systems. Digestion is associated by eating, the sensory system is taste. Withdrawal and analyze is associated with the sensory system smell, that is used to distinguish between good and wrong things: which can stay and which has to be thrown away, originally used in distinguishing the proper food. For physical activity, hunting, a good orientation - also visible orientation - is of importance. Sociability is guided by talking, which fits with phase fire. Hearing is associated with phase water, the discrimination between sleep and awake: a noise can wake you up. So each phase is associated with one of the five types of behavior and one of the five senses. This aspect is used in acupuncture diagnosis.

Now, behavior is described by the location of used muscles during each type of behavior (eat, standing, hunting). This follows the course of the yang meridians. Many acupuncture points of these yang meridians are located on the middle of the muscles. A good illustration are the locations of the yang meridian stomach and yin meridian spleen.
meridian on the belly. On the belly, points of the spleen meridian are located at the border of the muscles, in a kind of gutter. In terms of stiffness, noticing the amount of copper and zinc, reflecting the muscle activity: the yang meridian on the hill of the muscle notices this fast, and the yin meridian in the gutter notices this somewhat later, collecting the muscle activity information during a larger time scale. In this way, possibly the yin meridians have more copper, zinc and MT. This may explain why and acupuncture on certain yin meridian acupuncture points is effective on a larger time scale and may prevent side-effects caused by too less MT compared to the intensity of the therapy (duration of needle in the point).

**Details about how needling of an acupuncture point may influence an organ.**

How does the brain translate needling of an acupuncture point into an increase of the effectiveness of the immune system to cure (use self-healing properties) the organ? The vagus nerve communicates between organs and the brain. 90 percent of this connection is afferent and sensory. Sensory neurons of the vagus are located in the nodose ganglia. These neurons convey mechanical, chemical, osmotic thermal and possibly nociceptive information of organs to the brain. Tracing experiments demonstrate that the right and left nodose ganglia innervate (slightly) different peripheral sites [41]. According to copper and zinc hypotheses, a mechanical stimulus can also be the stiffness of the connection tissue around the organ. Section of the vagus nerve reveals a map of the body. How detailed this map is, is a question, but is assumed in this article. Many maps consists in the brain. It is practical to save neighbor information from senses to outputmap. (Kohonen type neural networks [43]) A particular behavior op a particular moment is directed to one goal, holding at that moment. All maps in the brain are possibly aligned in one common map in the brain. Origin of this common map can be: a map of goal directed eye movements, a map of the feet to walk to a particular direction associated to the goal, a map of the lower leg, a map of the hand to grab the goal, a map of the tongue to catch the food, or in the case of our prehistorical ancessters: to catch for instance a fly. Behavior is associated with organ (in)activity, for instance hunting and running is correlated with less peristaltics in intestines. In other words, the body map of organs of the vagus nerve is presumably also projected in this common map. Due to an local infection or local damage of the organ, hypothetically, a deviation on the body map of the vagus nerve is visible, which is projected to the common map. This local damage of the organ is projected back to the other maps like of the tongue (used in tongue diagnosis), of the pulse (used in pulse diagnosis), of feet (used in feed reflexology), etc. In most of the maps (e.g. tongue) this local deviation has no physiological function, but is a side effect of the efficient way of coding by the brain. However it is used by acupuncture as diagnostic tool. With respect to the nerve vagus: the other 10 percent efferent projection from vagus nerve to the organs, which is cholinergic, may have a function in the native inflammasome [41]. Due to viruses or other damage a cell decides to decrease its mitochondria, because a less efficient cell produce less viruses (in the case of viruses). This process is described in [42] as cell defense response (CDR). Cholinergic receptor agonists of vagus nerve stimulation significantly inhibits this native inflammasome activation [42] by a prevention of release of mitochondrial DNA [42, by the alpha 7 nicotinic acetylcholine receptor signal]. Acetylcholine attenuates mitochondrial damage [42]. Acetylcholine extracellular released, first accumulates in macrophage cytoplasm following ATP stimulation. This macrophage attacks the cell to put acetylcholine into the cell [42]. In the context of the copper and zinc hypotheses: due to an infection, a local change in the organ in copper may be a short
term increase due to loss of mitochondria, followed by a long term decrease, due to blood circulation. When this change of copper measured in the connection tissue by the vagus nerve, the vagus may project back: a decrease of acetylcholine to this damaged location. Reason of this: because in the case of viruses this cell should not be too active. Then the map of cholinergic projection may follow the sensorial map of the vagus nerve. The main issue of acupuncture is why needling of an acupuncture point may influence a certain organ. Possibly the needled acupuncture point makes a meridian information more flexible or healthier. This is followed by a healthier common map and a healthier body map of the vagus nerve in the brain. Then the vagus nerve stimulates the local damaged cell (cluster of cells) with acetylcholine, as if it is a normal peace of tissue without viruses. In case of damage without viruses, the cell cluster will become healthier. If viruses are active, one should be careful, because a healthier cell cluster may produces more viruses.

In summary, certain muscle movements influences certain acupuncture points, yielding a certain activity pattern on the map in the brain. In response to this the brain will plan a (following) future activity, an associated useful second behavior, combined with a release of hormones and neurotransmitters, and a change in respiration and blood pressure. This second behavior presumably show its own copper and zinc values. Because each acupuncture point is supported with blood circulation, again, all points theoretically notice this variation in copper and zinc plasma concentration. Different hormones and vegetative responses show possibly a correlation with copper and zinc values. Acupuncture may exert its working by manipulating or resetting this copper and zinc response. Chapter 6.1 described an acupuncture model, called wu xing in copper and zinc terms. The model contains five types of behavior with each its own copper and zinc distribution. Some behavior is correlated with (emphasis on) copper, some other behavior is correlated with (emphasis on) zinc. Before this description first, chapter 5 describes some physiological processes depended on zinc and/or copper.

5 GLOBAL PHYSIOLOGICAL INFLUENCES ON COPPER AND ZINC IN ACUPUNCTURE POINTS

A number of physiological items influences or correlates with copper and zinc in plasma will be described. Menstruation cycle influences copper and zinc. Temperature in the body and of the surround influences copper and zinc. Some diseases correlates with changes in copper and zinc. Some medicine influences zinc and copper. Zinc and copper show a circadian rhythm, and diet influences copper and zinc values.

5.1 Copper

Copper is related to initiation of movement. Physical work is associated with dopamine activity in the brain. Dopamine is present at the start of physical work, and involves initiative, making decisions, timing, for instance a decision to delay a start, and motivation. A chronic copper deficiency can lead to a deficiency of the neurotransmitter dopamine. Both the synthesis of L-DOPA and the decomposition of dopamine need enzymes with copper ions as a cofactor (tyrosinase hydroxylase, respectively beta-hydroxylase). A copper deficiency can lead to a worse timing of the amount of dopamine. Situations correlated with less dopamine, for instance rest, need a decrease in dopamine: this decreasing process (by beta-hydroxylase for decomposition of dopamine) may be too slow, resulting in too much dopamine. At other times, when initiation of movement is required, the increase
of dopamine may be too slow. Also the neurotransmitter norepinephrine is related to copper: synthesis velocity of catecholamines (neurotransmitter norepinephrine and dopamine) is dependent on the copper enzyme tyrosine hydroxylase.

5.2 Zinc

Zinc is important in digestion, respiration, immune system and sodium-resorption by kidneys. Digestion, start of immune system and respiration are related with zinc ions. In the digestion process, enzymes with zinc as a cofactor, are carboxypeptidases and aminopeptidases. Hedemann [44] measured in pigs that an increase of zinc in the diet promotes the activity of various digestive enzymes. Another important zinc-dependent hormone is insulin. Zinc plays a clear role in the synthesis, storage and secretion of insulin as well as conformational integrity of insulin [45]. Insulin may form a complex with zinc increasing the binding ability of insulin to its receptor. Zinc may stimulate insulin action and insulin receptor tyrosine kinase activity [46]). In a first stage of the digestion process, cells from salivary glands communicate with each other with the aid of zinc ions. A zinc deficiency correlates with a decrease in appetite. Because digestion enzymes are digested too, their cofactor zinc becomes available after digestion.

Stress lowers the production of acid and production of mucus in the stomach. The autonomic nervous system innervates the stomach. Inhibition of gastric acid secretion reduces zinc (and vitamin B12) absorption [48]. Zinc ions may stimulate stomach acid production by the zinc dependent carbonic anhydrase [47]. This enzyme transforms CO2 and H2O into H2CO3, a source of protons. By exchange with potassium, the proton is transferred to the stomach plasm. Acid prevents infection of the stomach by for instance Helicobacter. An infection by Helicobacter yields a decrease in absorption of vitamin B12 and zinc ions.

Another cell type in the stomach produces a buffer with mucus, to protect the stomach wall from damage due to gastric acid. Zinc ions stimulate this mucus synthesis by generation of heat shock proteins, enhancing mucoid protective ability of the stomach wall [49]. Less mucus may result in damage of the stomach by its acid, in stomach pain and distention. Acupuncture applied to the diagnosis 'heat in the stomach'', as will be described in next sections, may be described as a decrease in zinc'' uses in its formula point REN-12, located above the stomach [1]. Two points of zinc function are mentioned: a role in acid and in mucus production. If acupuncture really perform on this way its activity on the stomach of course must yet be demonstrated.

The exchange of zinc across the membrane is affected by aldosterone, other steroid hormones, including sex hormones and endogenous ouabain [49b].

In the immune system, zinc ions are involved. Zinc is active in T cells, prostaglandins, and macrophages. Zinc ions as cofactor are required by the synthesis of interleukin 2, which is used by T cells and prostaglandins for signaling. A zinc deficiency results in less efficient macrophages. Then more macrophages has to be produced to obtain the same effect. Macrophages attack antigens, e.g. foreign food. A zinc deficiency is indicated by a reduced resistance and skin problems [50]. The cytokine 'TNF alpha (tumor necrosis factor)' is produced by macrophages in systematic inflammation, and stimulates the acute phase. Kour [51] measured a reduction in inflammation of the skin, when he put galvanic zinc copper micro particles in the skin. The reduction of the inflammation was measured as a decrease in TNF alpha. Detailed research in a vitro culture of adipose tissue, shows that TNF alpha is most influenced by zinc, and in a lesser content by copper. When in this in vitro culture lipopolysaccharides are added, TNF alpha increases. The measure of increase of TNF alpha depends on the presence of copper and zinc in the culture. When zinc ions
are present in the gel of the in vitro culture, the increase of TNF alpha is smallest. With copper the increase is larger, and without copper and without zinc the increase of TNF is the biggest [52]. The amount of TNF Alpha is a measure of inflammation. In fact TNF alpha co-stimulates the expression of superoxide dismutase, an important antioxidant that catches the very toxic superoxide.

Epigenetics, inflammation and zinc. Zinc has anti-inflammatory properties. A decline in zinc status is observed with age [52c]. Methylation status of DNA of IL6 promoter region has shown correlation with IL6 gene regulation, and was associated with the pathogenesis of rheumatoid arthritis [52c]. Zinc deficiency resulted in progressive IL6 promoter demethylation [52c]. This could contribute to enhanced IL6 response [52c]. A chronic inflammatory state, in turn, can promote additional epigenetic alterations including aberrant DNA methylation, resulting in further dysregulation of zinc homeostasis by altering promoter methylation and expression of zinc transporters [52c].

Zinc and haemostasis.

Zinc plays a role in haemostasis and thrombosis [52b]. Hypozincemia leads to poor platelet aggregation and increased bleeding time [52b]. Activated platelets secrete zinc into the local microenvironment, and so the concentration of zinc increases in the vicinity of a thrombus [52b]. Zinc is an effector of coagulation, anticoagulation and fibrinolysis [52b]. A review of these mechanisms is given in [52b]. TCM: Emptiness in Spleen, which may be related with hypozincemia, can result in easiness of receiving blue spots on the skin by bumping. In fact, zinc deficiency may be related with a proposition to bleeding. A glycoprotein in the blood, histidine-rich glycoprotein (HRG) promotes coagulation [26c]. Heparin is an anticoagulant. HRG may bind to this heparin, which promotes coagulation [26c]. The formation of HRG-heparin complex is enhanced in the presence of zinc [26c].

The role of MT in controlling Zn2+ homeostasis appears to be important in innate immune cells. In macrophages, MT deficiency impairs cytokine production and bactericidal activity after lipopolysaccharide (LPS) stimulation as a result of Zn2+- deficiency [129].

Acupuncture at ST36 on the leg decreases the increase in TNF alpha in the COPD rat model in the lungs [53]. Acupuncture at ST36 and GV20 suppress the expression of TNF alpha proteins in bilateral ischemia and non-ischemia cerebral regions in rats [54].

Respiration, with respect to shortness of breath, has the following relation with zinc ions. Respiration is not only controlled by the need of oxygen, but in fact more stringently controlled by the amount of CO₂. More CO₂ indicates a shortening of breath. Zinc ions have a function in the homeostasis of a constant acidity level in blood. This is shown by the zinc dependent enzyme carbonic anhydrase, which captures CO₂ from the blood and converts this into bicarbonates. In the lungs, carbonic anhydrase converts bicarbonate back into CO₂, where it can be breathed out. Without this enzyme, or possibly when zinc decreases, the acidity level easier leads to shortness of breath. Furthermore, zinc stimulates de novo lipogenesis in alveolar cells to produce surfactant, a surface active lipoprotein, for optimal working of the lungs [12].

De novo lipogenesis uses citrate in the Krebs cycle [5, 6]. De novo lipogenesis is stimulated by zinc, by the following process: Citrate becomes free because zinc inhibits the citrate oxidation (by inhibiting the activity of m-aconitase). Citrate is also used in de novo cholesterol biosynthesis, which occurs in glands to produce steroidhormones. In this way,
zinc - which releases citrate by the inhibition of citrate oxidation - with menstruation cycle and fertility.

In the kidneys the enzyme zinc metallopeptidase (Neutral endopeptidase, NEP) is expressed. It decreases the backresorption of sodium by kidneys [55]. This is done by inactivation of enzymes that are responsible for sodium backresorption (atrial natriuretic peptide, bradykinin and angiotensin 1). Zinc metallopeptidase contributes to prevention of hypertension. The occurrence of more zinc may lead to more frequent urinating, a characteristic used in acupuncture diagnostics.

Many eye problems are decreased by zinc ions [35]: macular degeneration can improve when zinc is administered, and zinc deficiency can result in conjunctivitis and night blindness. Eyes need much zinc: forty percent of the total amount of intracellular zinc is present in eye and prostate. During vision a number of zinc-dependent enzymes are active in the eyes: retinoldehydrogenase, alpha-mannosidase (lysosome enzyme), carbonic anhydrase (cilear body), corneal collagenase (lens), leucine aminopeptidase [35].

5.3 Menstruation pattern correlates with a rhythm of copper and zinc fluctuation.

Acupuncture diagnosis involves the menstruation pattern. The menstruation pattern is correlated with a monthly, copper and zinc fluctuation in plasma. Michos [56] measured that plasma concentration of zinc ions correlates positively with the amount of estradiol. Furthermore, he found that plasma concentration of copper ions correlates negatively with the amount of estradiol. This means that a fluctuation of copper and zinc is correlated with the monthly (menstruation related) fluctuation of estradiol. After menstruation, estradiol increases, leading to an increase of zinc. After ovulation a decrease in estradiol takes place. This leads to an increase in copper ions. This illustrates that menstruation pattern contributes to the diagnosis used in acupuncture.

5.4 Heat and cold in the environment.

Heat and cold in the environment, seasonal influence, in principle challenges the maintenance of body temperature. The regulator of temperature, the thermostat is a function of hypothalamus. But warmth is generated by activity of the muscles. The most important warmth generator, independent of muscle activity is brown adipose tissue, located in the upper back. They perform this with a particular deviation in their ATP generation. These adipocytes contain in their mitochondria a unique "uncoupling protein 1" which uncouples complex IV and complex V by inducing the H+ gradient independently from ATPase, whereby the energy dissipates as heat, instead of production of ATP [57, 58]. The heat is transported by circulation through the body. Copper is a cofactor in complex IV, and zinc influences the prior complex III in the mitochondria [15], thus, copper and zinc manipulates the heat generated by the brown adipose tissue. They may regulate a gain in temperature. The acupuncture point dumai 14 located on the upper back near the brown adipose tissue shows heat regulating properties, and is involved in treatments of fever, heat or cold signs [1]).
5.5 Copper and zinc dependency in some diseases.

An influence of acupuncture on diseases may be exerted via the copper and zinc dependency of these diseases. Then these diseases may influence the stiffness of some acupuncture points. The following illnesses are accompanied by an increase of copper ions: migraine, depression, epilepsy, stomach ulcers, inflammatory bowel disease, Parkinson disease, aneurysm, diabetics and obesity. Dhillon [27] added zinc in the diet of a group of migraine patients. In response, the migraines disappeared. Zhou [28] measured in rats (stressed or depressed) that the amount of copper ions was increased, compared to normal, and that the amount of zinc ions was decreased. When these rats were given electro acupuncture, a decrease of copper ions and an increase of zinc ions was induced. Consumption of the antidepressant maprotilin in these rats shows the same effect: a decrease of copper and increase of zinc. Doretto [60] measured in animals with epilepsy a decrease of zinc and an increase of copper in hair and in serum. An ulcus in the stomach results in an increase of copper in the serum [12]. Inflammatory bowel disease correlates with a deficiency of zinc [61, 62]. In fibromyalgia a deficiency in zinc is measured [63].

Parkinson disease (PD) is determined by a decrease of dopamine, produced by a decreased amount of cells in the substantia nigra in the brain. PD may be correlated with a less efficient copper ion transport, because PD can be associated with a hereditary less efficient ceruloplasmin, a transporter of copper ions in the blood [64]. When copper is less transported, copper ions will accumulate in the brain. Because the substantia nigra possesses much copper [60], it is vulnerable for oxidative stress. Cells of the substantia nigra die. One of the first symptoms preceding PD is a decrease of smell, which correlates with a decrease in zinc ions, and may be caused by the mentioned increase of copper ions. Different research is done on the relation between acupuncture and PD. Chapter 6.1 describes PD related to copper or zinc, depending on the type of acupuncture point. Of course, the degenerative disease PD is not reversible, and exact cause is not known, but points for instance to certain agriculture pesticides and possibly a role of advanced glycation end products, influenced by diet [98].

A copper deficiency is measured in patients with an aneurysm of the aorta and in patients with cholesterolasma [67].

Hypothyroidism is related to zinc: the synthesis of thyroid hormones requires zinc and copper, and conversely, thyroid hormones are essential for the absorption of zinc [68].

Diabetics often show a deficiency in zinc [45]. Garcia, [69] measured in obese people, that obesity is correlated with a deficiency of zinc. This may be related with the finding that zinc and fatty acids compete with each other on albumins, their transport protein in blood [70]. Acupuncture positively influences diabetes [71] and obesity [72].

Although smoking is not a disease, it is interesting that cigarette smokers have a significant higher serum copper concentration [73b].

5.6 Acupuncture and Western medicine

Big amounts of paracetamol (acetaminophen), an anti-inflammatory medicine decrease the effect of acupuncture. This can be explained in term of the local increase of zinc after needling. With respect to paracetamol: acupuncture causes a release of glutathione [18]. This glutathione releases zinc ions from MT [19]. While in the liver, the metabolite of paracetamol decreases glutathione [73] and therefore may decrease the effect of acupuncture, by a decrease of by acupuncture increased amounts of zinc.

Other medicines like the contraceptive pill increases copper in blood, and decreases zinc. Drugs against hypotension, such as indapamide and amlodipine lower zinc and copper levels [20]. A side effect of zinc deficiency is glucose intolerance and insulin resistance.
A side effect of copper deficiency is hypercholesterolemia [20]. These side effects of anti hypertension medicine are contra productive. Adding copper and zinc to these medicine improves the anti hypertension effect [20].

Drugs that produce a favorable effect on zinc metabolism are, those inhibiting angiotensin-converting enzyme activity, spironolactone, substances that rise sensibility of tissues to insulin and insulin itself (but also weight reduction profitably influences zinc metabolism) [49b].

5.7 Circadian rhythm in copper and zinc ions.

The availability of copper and zinc during the day is not always the same, due to a hypothetical circadian rhythm in copper and zinc ions. Acupuncture also describes that certain symptoms (and behavior) prefer particular times of the day. TCM called this the Zi Wu cycle. A regular measurement of Vallee [35] indicated that the zinc concentration in the serum in the morning is higher than in the afternoon. In fact, MT is merely an intracellular protein, and the circadian rhythm of zinc is measured in the serum. Also [73b] measured a circadian rhythm in serum zinc concentration, decreasing from early morning to late afternoon. This may be due to a reduction in serum zinc 60 min after a meal (glucose) [73b]. On the contrary they found that serum copper did not show a circadian rhythm [73b]. The only reliable indicator of copper status in the amount of copper in the liver [73b]. Because of the reactivity of copper, liver may adopt (store) the increase of copper during the circadian rhythm.

Theoretically, because copper and zinc compete in MT, the circadian rhythm in zinc should be associated with an opposite circadian rhythm in copper (possibly only felt in the liver). In fact the liver may direct this competition somehow (see chapter EPF), by producing a situation dependent amount of transport proteins of copper and another amount of those of zinc respectively (ceruloplasmin respectively albumin).

How is this circadian rhythm in copper physiologically regulated? During the day, a period of physical work, much energy is needed. ATP is the molecule of energy and copper plays a role in ATP formation, so an increase of copper is required. The increase of copper is used to optimize dopamine. Dopamine is a neurotransmitter related to motivation, initiation and timing of movement. Both production and removal of dopamine require copper ions. On the other hand, during sleep, less copper ions are needed. Decrease in copper. During sleep, the liver collects blood and extracts copper from this. In the evening: during the fall of daylight, the pineal gland in the brain produces melatonin. Melatonin binds the copper ions, presumably takes it away from other brain processes, resulting in a reduction of copper ions in the brain [74]. Acupuncture results in an increase of melatonin [75]. Melatonin crosses the blood brain barrier [75b]. Copper ions leave the brain probably by the brain liquor circulation. The function of this circulation, is to maintain a biochemical stable brain. The liquor is produced in the ventricles of the brain, flows to the base of the brain enters the arachnoidal space and flows to the vertex. The liquor is resorbed back into the venous blood system at the vertex, which is also the location of a strong acupuncture point, dumai 20, and which is located on a so called internal branch of the liver meridian (wood). This point of back resorption contains much MT, regular assumed for detoxification, and according to this article may be also used for regulation of copper and zinc. On the base of the brain, the important acupuncture point’s dumai 16 and
gall bladder 20 located. These acupuncture points may influence the flow of copper, by needling. Furthermore, longer sleep duration correlates with a higher zinc ion concentration (and zinc/copper ratio) in serum and hair [76].

**Increase in copper.** The gastrointestinal tract receives copper from the diet. Most of the copper in the gastrointestinal tract originates from saliva, bile, gastric secretion and apoptotic intestinal cells [29]. Intestinal pH (acid environment) is probably the most important physiological factor affecting copper absorption [29]. By portal vein, the liver receives copper ions. The copper uptake by hepatocytes is stimulated by the reducing agent vitamin C that probably speeds up copper reduction (from Cu2+ to Cu+) [29].

Synthesis of ceruloplasmin (that transport copper in blood) in liver is copper dependent [29]. Another way of increase in copper may be by cholecystokinin released by the nervus vague. This hormone stimulates the release of bile from the gall bladder.

To prepare for a period of physical activity, epinephrine and thyroid gland hormones plays a role in the process to increase copper ions. A third role is played by the enterohepatic circulation (EHC). (1) The liver activates thyroid hormones, in which T4 is converted into T3. A decrease in zinc ions in the liver initiates the activation of thyroid gland hormones by the liver [77, 78]. This decrease in zinc ions is for instance noticed by the liver via the portal vein, which carries zinc ions resorbed from the intestines (dinner, and zinc form zinc-dependent digestion enzymes). T3 crosses the blood brain barrier and increases the number of dopamine receptors in certain parts of the brain [79]. This is a preparation of the next phase, physical work, in which dopamine plays a role. (2) Epinephrine stimulates the release of copper ions in the liver by stimulation of synthesis of ceruloplasmin, an intercellular transport enzyme of copper. This spreads copper in the body to all cells via the blood system [26]. (3) Increase of copper can take place by secretion of bile from the gallbladder. According to the EHC, these copper ions are reabsorbed from the intestine into the bloodstream and reach the liver via the portal vein.

Together with this variation, an increase of bile acids are absorbed from the intestines into the body. Then, in the periphery of the body, the brown adipose tissue and muscle increases energy expenditure. This is performed indirectly, by increasing the pathway of intracellular effect of thyroid hormones [81]. Thus, the EHC may regulate an increase of energy expenditure, together with regulation of a temporarily increase of copper. Chapter 5.8 describes the effect of diet on bile acids and the effect of a decrease in bile acids on metabolic syndrome, the (auto-) immune system, and dermatological complaints.

**Qi stagnation.**

A circadian rhythm in copper and zinc ions is the reason that a proper transport of copper and zinc is essential. Some flow, transport in copper and zinc is needed, to let enzymes with these ions as cofactor work most efficient. This flow may be stagnated. In acupuncture, Qi stagnation is related to liver and gallbladder. In the above described circadian rhythm in copper, the liver and EHC play a main regulating role. Acupuncture may improve this flow. In hamsters with cholelithiasis, acupuncture on gb34 and lv14 both on the right side increases secretion of cholic acid [82]. Chapter 7 describes a role of glutamate receptors reacting on a decrease in copper and zinc ions.
5.8 Diet, carbohydrates, baked and fried fat.

Too much carbohydrates (for instance sugar) contribute to diabetes type 2, hepatic steatosis, an important intermediate stage of diabetes type 2, and can deteriorate to liver fibrosis, by different ways: (1) much carbohydrates in the diet, and specific glucose and fructose increased the change of advanced glycation end products (AGE) in obesity [75e]. AGE is built by misfolded proteins, that initiate neuro degeneration processes, like Parkinson’s disease and Alzheimer’s disease [98, 99]. Another contribution of liver fibrosis are (2) baked and fried fat: bad for the liver: rats fed with a diet of glycolytinexes acquired via baking show an increase of liver fibrosis [84].

We will describe how obesity may depend on copper and zinc deficiency. A zinc deficiency may contribute to obesity: (1) Administration of a substantial amount of zinc ions in the diet stimulate the recovery from hepatic steatosis [85], one of the symptoms of obesity. (2) Another argument is the correlation between obesity and inflammation characteristics in adipose tissue [86], like TNF alpha (tumor necrosis factor). Zinc inhibit the increase of TNF alpha (vitro culture) [51].

A copper deficiency may contribute to obesity:

(1) Administration of copper increases the EHC [87]. Mice exposed to high doses of copper, show a decrease in body weight, and an increase of total bile acid level [88]. Possibly obesity is correlated with a decrease in EHC. The following reasoning is given:

Diet carbohydrate and fat influences the composition of gut micro biota [86], and thus the composition of bile acid, because these microbiota changes a fraction of bile acids into the so-called secondary bile acids. Via EHC, bile acids circulate throughout the body, in skeletal muscles and adipose brown tissue. Certain types of bile acids - receptors (FXR) are related with glucose intolerance, which contributes to obesity. Other types of bile acids (secondary bile acids) and their receptors (TGR5) in brown adipose tissue and muscle may prevent obesity by increasing energy expenditure, which is performed by increasing the efficiency of intracellular pathway of thyroid hormones [81]. More energy expenditure results in less stored energy contributing to obesity. These healthier secondary bile acids depends on microbiota in the gut, and the gut microbiota depends on diet. This is why chronic too much sugar in the diet causes obesity and metabolic syndrome.

Bile acids can regulate triglyceride, cholesterol, energy and glucose homeostasis [84]. Bile acids have been reported to inhibit diet induced obesity [89] and prevent the development of insulin resistance (in the beginning of DM2, not in a later stage) [84].

(2) The metabolic syndrome is diagnosed by a co-occurrence of three out of five of the following medical conditions: abdominal (central) obesity, hepatic steatosis, elevated blood pressure, elevated fasting plasma glucose, high serum triglycerides, and low high-density cholesterol (HDL) levels. Obesity correlates with cholesterolemia. Klevay added copper in the diet, which result in a decrease of cholesterol in the blood [67]. Bile acid synthesis needs cholesterol, and less cholesterol remains in the plasma.

Contribution of albumin to diabetes mellitus. Albumin is involved in a proper blood circulation, by its contribution to osmotic pressure. A decrease in albumin may result in edema. Insulin is involved in albumin synthesis by the liver. Insulin is required for adequate albumin synthesis: diabetics have a decreased rate of albumin synthesis [26c]. A decrease in available insulin (insulin resistance) results in a decrease in albumin, and a proposition tot edema. Less albumin may result in less circulating zinc in the blood [70]. In turn, less zinc have a negative influence on insulin receptors. In this context, it is useful to prevent too much zinc loss, to optimize insulin receptors, thus to optimize albumin...
synthesis, thus to prevent edema. Less albumin may correlate with tiredness: albumin transports fatty acids, which are the primary source of energy for many body tissues, such as resting skeletal muscle, renal cortex, liver and myocardium [26c]. Another aspect of less albumin is less transport of secondary bile acids, used for increase of energy expenditure, preventing obesity.

Furthermore, an aspect of binding of fatty acids to albumin, is the effect of this binding: a change in conformation of albumin (allosteric effect) [26c, 33b, 89b], which may result in a worse binding of other components to albumin, like hormones, steroids, thyroxine, bile acids, vitamin B12 and D, leptin, heme, bilirubin, and pharmaceuticals (like barbuturates and ibuprofen) and minerals (calcium, sodium, potassium, magnesium [33c]). It is not known if this conformation change also influence zinc binding to albumin. The decrease in transport of thyroxin may contribute to tiredness (although a feedback mechanism by hypophyse may compensate this by stimulation of the thyroid function). Another aspect is the decrease of transported leptin by albumin, increasing appetite. Leptin, the "satiety hormone", is a hormone made by adipose cells that helps to regulate energy balance by inhibiting hunger. The increase in hunger may contribute to obesity. In this hypothetical context, too much fatty acids is a risk for obesity, by different mechanisms.

Regulating mechanisms with respect to the amount of albumin in the blood takes place in the liver. Diverse hormones, like insulin, glucagon, thyroid hormone and growth hormone regulate albumin synthesis in the liver [86b]. This synthesis is carefully regulated, to optimize blood circulation and delivery of nutrients [86b]. More glucose in blood (for instance during growth) influences colloidal pressure, and liver increase albumin synthesis. Because zinc is mainly transported by albumin, hypoalbumenia may decrease or slow this transport. A risk of edema in this context is correlated with hypothyreoida, and diabetes mellitus. Finally, if the liver is not capable to produce albumin (malnutrition, hepatosteatosis) overrules this subtle regulation, to create hypoalbumenia.

**Summary of chapter 5.**

In summary, the stiffness of an acupuncture point is influenced by: (1) recent muscle activity or inactivity in the neighborhood of the point. In addition, Cu and Zn in the blood circulation around the acupuncture point, influences its stiffness; by (2) behavior; (3) menstruation pattern; (4) heat and cold in the environment; (5) some disorders; (6) some regular medicine; (7) circadian rhythm, and (8) diet. This explains why an acupuncture treatment differs between two persons with for instance headache. It is only equal, if the two persons show the same copper and zinc pattern: same behavior type, same circumstances, same other complaints, reflected in their acupuncture diagnosis.
6. TCM terms translated in copper and zinc, hypothetical.

6.1 The acupuncture concept Wu Xing in expressed in copper and zinc.

In the brain, diverse associations are made between the map of acupuncture points and physiology and behavior. One candidate can be a scheme used in acupuncture is the Wu Xing model, which consists of five phases: water, wood, fire, earth, and metal. In the model of Wu Xing, each phase correlates with certain characteristics and symptoms of the patient. Chapter 4 describes meridians as a correlation between certain movements with five types of behavior and the five senses. According to Wu Xing, each meridian belongs to one of the five phases of Wu Xing. The Wu Xing model can be expressed in copper and zinc terms. The five phases are aligned according a cycle, one feeding the next, influencing in terms of energy, expressed in needle formula. Earth feeds metal, metal feeds water, water feeds wood, wood feeds fire, and fire makes the circle round, by feeding water. Phase wood is responsible for copper dependent processes, phases earth and metal are responsible for zinc dependent processes. These processes al already described in chapter 5. Table 1 summarizes for each of the phases the hypothetical ions.

<table>
<thead>
<tr>
<th>Phase in Wu Xing</th>
<th>Hypothetic ions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Water</td>
<td>Mg (Zn and Cu) regulation</td>
</tr>
<tr>
<td>Wood</td>
<td>Cu &gt; Ca &gt;</td>
</tr>
<tr>
<td>Fire</td>
<td>Mg &gt; Ca &lt;</td>
</tr>
<tr>
<td>Earth</td>
<td>Zn &gt; Ca &gt;</td>
</tr>
<tr>
<td>Metal</td>
<td>Mg &gt; Ca &lt;</td>
</tr>
</tbody>
</table>

Table 1: hypothetical ion behavior for each of the phases in Wu Xing. Magnesium (mg) is involved in phosphorylation processes, copper is involved in regulation of certain neurotransmitters (catecholamines) and zinc is involved in glucose uptake in each cell.

Phases fire and water.

Phases Fire and Water regulate the transition from a dominance of copper processes, into dominance of zinc processes and vice versa. They perform this by decreasing the intracellular concentration of calcium ions, to generate a kind of pause. This pause is generated as follow in detail: an intracellular decrease of calcium ions, is performed by (a decrease of) several hormones (correlated with certain behavior changes) and leads to a decrease in the DNA expression. This results in a relative decrease of certain enzymes (used in the previous phase). The increase number of free amino acids will be used in the next phase for other enzymes in other physiological processes. In the (relative) pause of DNA expression, a change of choice of expressed enzymes can occur. On this way, one amino acid can play a role in different enzymes, in both phase earth and phase wood at distinct times of the day. During digestion, other enzymes are active (enzymes with zinc as cofactor), and another part of the DNA is expressed than during physical work (enzymes with copper as cofactor). After a decrease in DNA expression of enzymes in phase earth,
there can be a reset in locations of active DNA expression.

Phase earth needs an increase of calcium: after digestion, zinc transport is needed for insulin receptors to transport glucose into cells. In active zinc transport, Calcium is needed [1b]. Phase wood needs an increase of calcium for DNA expression of copper related processes, for instance with respect to dopamine metabolism.

Phases water and fire separates phases wood and earth, also separating these phases with respect to each other. Phases water and fire are related to the autonomic nervous system, and are initiated by certain behavior. Behavior with less activity: sleeping, sitting causes a decrease in certain signal molecules (epinephrine, dopamine), and thus less intracellular calcium. During physical work, heart rate is faster than during rest. In the pause after physical work, intention is that the heart rate decrease. Phase fire is responsible for this change. This is a decision of the autonomic system. Calcium contributes: the impulse conduction of the muscles of the heart makes use of calcium. Too much calcium results in cardiac arrhythmia. A decrease in calcium will contribute to a decrease in heart rate. The fact that in mice red blood cells and bones are enriched with zinc [101] may indicate the transport function of phase fire (associated with heart and vessels) and the storage function of phase water (associated with bones), both with respect to zinc. During phase water and fire, calcium is supposed to decrease, and no extra protein synthesis is supposed to occur: only transport of zinc (fire) and storage of zinc (water) takes place. According to Wu Xing, one type of disturbance of phase water, is associated with night urinating. By urinating (and sweating), relative more zinc leaves the body. These processes do not need much change in enzymes, and are apparently correlated with phase water, and is associated with an optimum in zinc. When during maximum zinc period in the night, urinating takes place, enzyme NEP does not contain much zinc of less enzyme of it is present. Biochemical details: In the transition from copper dominance to zinc dominance, how does an increase of zinc cause an increase of calcium? After the release of zinc from MT causing a zinc increase, this zinc is bonded to zinc transporter. Zinc transporters regulate a strong intracellular gradient, ranging from pico to micromolar concentration of zinc. A subtype, zinctransporter1, binds to a (L type) calcium channel, to increase the calcium influx into the cell [102]. Then phase earth is present with increase in zinc and calcium.

**Acupuncture on meridians of phase wood.**
The following research demonstrate a relation between phase wood and copper, respectively a relation between phase water and zinc. Acupuncture using an acupuncture point on a meridian of phase wood may differ from using a meridian of phase water. The acupuncture points LR3 and GB34 are located on meridians that belong to phase wood. Needling these points in the Parkinsonism animal model improves motor dysfunction and inhibits the reduction of tyrosine hydroxylase in the nigrostriatal dopaminergic neurons [103]. This enzyme transforms tyrosine to L-dopa and determines the rate within the synthesis of catecholamines (dopamine, norepinephrine). This can be related to copper: other research illustrates that a copper deficient diet results in a low tyrosine hydroxylase activity [104].

**Competition between phases of wu xing.**
In Wu Xing mutual competition exist between phases wood and earth and between phase's wood and metal (between the yin meridians [103]). This competition can be used in certain needle formula. For instance, an imbalance in earth can be treated in acupuncture, by needling of points associated with phase wood. The fact that copper and
zinc ions compete on the binding site of their intracellular transport protein metallothionein can describe this competition.

**MT as sensor of phase of wu xing.** Other regular research, research of messenger DNA of MT, can be related to the supposed role of MT in Wu Xing. The five phases of Wu Xing are each associated with an emotion: roughly spoken: phase metal with sorrow, phase water with fear, phase wood with anger, and phase earth with worrying, mulling. In the hypothetical Wu Xing description, MT intracellular shows the phase of Wu Xing by the ratio of its bounded copper and zinc. So MT may contain intracellular information about the actual, recent or chronic emotional state. This hypothesis can be related to finding that increase of MT may occur in addictions. Several stimulants, amphetamines, smoking and alcohol increase the gene expression of MT [105, 106]. With the increase of MT, emotions are intracellular better observed. In this context, the finding that lithium, a medicine for manic depression, decreases messengerRNA of MT [107] can be an explanation for its side effect, flattening of emotions.

6.2 Some other terms used in acupuncture:

6.2.1 Backshu points related to zinc

The Acupuncture points GV14, GV20 are located on the du mai meridian which enters the kidneys (1). The kidneys belong to phase water. Needling of these points in the parkinsonism animal model (rat) results in a decrease of the abnormally elevated glutamate levels [122]. This can be described by the fact that acupuncture release zinc ions, which can slow down the signal transfer of receptors of glutamate. Interesting in relation to the bladder meridian and du mai meridian located on the spine, is the occurrence of an accumulation of zinc vesicles at inhibitory terminals in the spinal cord, in the dorsal and ventral horns. Here, zinc may act as modulator at mixed inhibitory synapses: synapses with a co release of GABA and glycine [123, 124]. Important acupuncture points are also located on the bladder meridian are, namely the so-called backshu points. As is described, water may be related to (an optimum of) zinc, as a passive store: during the circadian rhythm, in the night and morning.

6.2.2 Shu Transporting Points, related to metallothionein

A special type of acupuncture points on each meridian, the shu transporting points can now be expressed in terms of MT. These points are located on the lower arm and lower leg course of the meridian. Lower arms and lower legs are closely involved in behavior, and play a role in the association by the brain with certain behavior. Shu transporting points show characteristics of the meridian on which they are located, added with characteristics of one of the five phases of Wu Xing. Needling of a (yin) shu transporting point at the fingernail, respectively toe nail, influences wood properties, the next proximal shu transporting point influences fire properties, the next point earth properties, the following point metal properties and on the elbow or knee, the shu transporting point has water properties. When we translate this sequence into copper terms: wood is correlated with maximum copper, fire regulates a decrease in copper, earth is neutral (zinc in digestion system), metal is associated with an increase of zinc outside the digestion system, and water occurs at rest.

The circadian rhythm of copper describes during phase water, the liver collects copper, to obtain a maximum zinc concentration in the brain. Toe and finger are relative cold parts of our body. To compensate for this lowering of body temperature, to keep the location
warm, more energy (ATP) is needed. Copper ions accelerate the availability of ATP. Nails, the coldest location, needs the biggest amount of copper. Because we suppose that each cell "obeys Wu Xing", this means that more MT should theoretical be present in the acupuncture point at this nail, with a proper ratio of copper and zinc, fitting the time of the day and behavior according to Wu Xing.

Application of shu transporting points in acupuncture: An application how the neural map in the brain of activity pattern of all acupuncture points is read by the brain is the following. Certain acupuncture points, the shu transporting points can be used in a formula, describing interactions between different phases of Wu Xing (feeding and controlling mechanisms, according to the Sheng and Ke cycle). The 4 needle technique according to Hicks describes which points should be chosen in the needle formula [125]], using the five elements theory and Wu Xing dynamics. The local temperature in shu transporting points is determined by the temperature of the surround and the relative distance on the extremity to the end of the extremity (tow, fingertip). Also circadian rhythm estimates the change on a certain body temperature. This body temperature changes copper/zinc ratios of shu transporting points. To ignore the influence of the surround, point on about the same location on the extremities are correlated with each other, to get an idea of phases of Wu Xing and its feeding and controlling mechanism. This leads to certain needle combinations on same extremity position.

On this way, the effect of the surround temperature is excluded in the mechanism on the map in the brain of acupuncture point, to read the situation that a feeding moment occurs (e.g. phase metal feeds water) or a correction moment occurs (e.g. phase earth controls water).

6.2.3 Yin is related to zinc and Yang is related to copper ions.

Yin and Yang are two central terms used in acupuncture, and can be assigned to many phenomena: for instance, an organism contains many feedbacks, oscillations, which can also be expressed in Yin and Yang. In the context of our article, we consider one aspect of yin and yang: we will address copper and zinc to Yin and Yang. Yin is associated with storage and zinc ions. Zinc, for instance related to de novo lipogenesis and de novo cholesterol synthesis, can be considered as Yin. Zinc is used in the uptake of glucose in the cell. Production of ATP from glucose is inhibited by zinc ions. Yang is associated with activity and copper ions. Activity needs ATP, the universal intracellular source of energy. ATP production is stimulated by copper ions. Initiation of movement is related with dopamine. Yin emptiness and yang emptiness. Zinc is Yin, copper is Yang. It seems contra intuitive that yin emptiness is connected by copper instead of zinc, and yang emptiness is connected by zinc instead of copper. Zinc and copper compete. Yang emptiness means a relative increase in yin.

6.2.4 Yang emptiness is related to zinc deficiency.

In acupuncture theory, yang emptiness is related to cold sensation and polyuria. Hypoglycemia leads to a cold sensation. By urinating, zinc leaves the body, so polyuria may induce zinc deficiency. Furthermore, in the kidneys, a decreased effective zinc
dependent enzyme NEP is relevant in yang emptiness of kidneys, with less zinc ions and polyuria. A close interaction between thyroid hormones and zinc exists [127]: Zinc is required for the T3 receptor: zinc supplementation improved thyroid function in hypothyroidea patients with low zinc levels. Otherwise, thyroid hormones are essential for the absorption of zinc.

**Yin emptiness and Blood emptiness are related to copper deficiency.**
Copper is after iron and zinc the third most abundant essential transition metal in human liver [31]. To describe yin emptiness and blood emptiness in copper terms, we include iron ions. Blood emptiness starts with loss of blood: loss of iron ions. Yin emptiness (a relative chronic character compared to blood emptiness) starts with copper deficiency, which indirectly causes an iron deficiency.

### 6.2.5 Yin emptiness: copper deficiency causes iron deficiency.

Chapter 6.2.5 describes the role of copper and iron in yin emptiness, and jet ignores the contribution of magnesium. After description of role of magnesium in Chapter 7, Chapter 7.10.4 includes magnesium deficiency in Yin emptiness.

Yin emptiness is a worse condition than blood emptiness and may start with copper deficiency. Copper deficiency causes iron deficiency by the decrease of the protein ceruloplasmin. The copper-containing ferroxidase ceruloplasmin plays an important role in iron homeostasis and thus links copper and iron metabolism [31]. Ceruloplasmin transports copper in blood. A second task of ceruloplasmin is that it captured Fe$^{2+}$ and oxidizes it by Cu$^{2+}$ to Fe$^{3+}$. Dietary and recycled iron ions are in the Fe$^{2+}$ oxidation state. Fe$^{3+}$ is the form which is transported in serum by transferrin [131]. So a copper deficiency, and thus a deficiency in ceruloplasmin yields a deficiency in iron.

**Empty Heat.**
Yin emptiness can turn into "yin emptiness with empty heat". A sign of empty heat is a heat sensation during late afternoon (around 17 o'clock). Another sign is "heat of the five palms" which means relative warm hands, feet and chest. How do copper and zinc correlate with this? Yin emptiness is a deficiency in copper. Chapter 5.4 describes a relation between copper and body temperature generation by acceleration of ATP synthesis in the mitochondria. If this diminishes, The hypothalamus as thermostat increases the temperature generation. The two signs of empty heat show the copper distribution in the body surface in location and in time. According to the circadian rhythm, the copper optimum is in the afternoon. According to the characteristics of shu transporting points, MT is maximal at the extremities, hands and feet. The fifth "palm" on the breast may be influenced by the brown fat tissue on the back between the shoulders. More MT is in most of the cases more local copper.
6.2.6 Blood emptiness:
First a relation between blood emptiness and vitamin B12 and iron will be described. Second, the relation between blood emptiness and copper (yin emptiness) will be described. Third, the relation between blood emptiness and zinc (phase earth, spleen meridian as a therapeutic way in TCM to treat blood emptiness) will be described.

(1) Relation between blood emptiness and vitamin B12.
Temporary severe blood loss leads to symptoms of blood emptiness. Blood emptiness has probably a relatively reversible character (compared to yin emptiness), for instance it can occur temporarily after a heavy menstruation or other severe blood loss or a vitamin B12 deficiency. Copper is important to let the blood emptiness vanish: by increasing ceruloplasmin; by upregulation of expression of DMT1 (dication metal transporter 1, [138]), by upregulation of ferroxidase hephaestin, and upregulation of the iron transporter ferroportin1. This leads to hypothesize that a vulnerability to obtain blood emptiness increases in circumstances of low copper.

The brain tries - in case of iron deficiency - to upregulate copper by a process in the blood brain barrier:

Brain copper is derived from peripheral copper that is transported across the blood-brain barrier (BBB), which separate the brain interstitial space from blood and cerebrospinal fluid (CSF), respectively [22d]. The BBB represents the major route for the transport of copper from the blood circulation into the brain parenchyma, where copper is utilized and subsequently released into the CSF via the brain interstitial fluid. The copper in the CSF can be taken up by choroid epithelial cells, from where it may be stored or exported to the blood [31]. (The choroid plexus is located in the brain ventricles, and is an important factor of the blood brain barrier. The choroid plexus produces cerebrospinal fluid and protects for heavy metals.)

Anemia increases the copper level in the blood brain barrier [128, 131]. Monnot measured in cells of the choroid plexus (in BCB) a diminishing expression of cellular surface transporters (DMT1 (particular) and Crt1). This means a decreased cu uptake, and a reduction in cu exporters (ATOX1, ATP7A), and thus an increased cellular retention of cu in the brain. Iron deficiency led to significant increases in Cu levels in brain parenchyma and the choroid plexus.

(2) relation between blood emptiness (vitamin B12) and copper (yin emptiness).

Vitamin B12 is an important vitamin which influences the stability of the mind, expressed in a presence of calmness, which is in acupuncture addressed as a disharmony in the energy of the heart meridian. Adding B12 can diminish a depression. A feature of both blood emptiness and yin emptiness is dryness. This is visible at the level of the tongue. Also vitamin B12 deficiency is shown by glossitis [142] a serious dry tongue. Copper deficiency shows similarity with vitamin B12 deficiency [132]. One cause of myelopathy is vitamin B12 deficiency [133]. Severe copper deficiency is also a cause of peripheral neuropathies [133]. Synthesis of the phospholipids in myelin is done by the copper dependent cytochrome c oxidase [108]. Severe copper deficiency is a cause of reversible refractory anemia. So copper and B12 are closely related.
How is B12 related with copper? A decrease in the one seems to cause a decrease in the other. Copper deficiency may influence vitamin B12 adsorption. Extracellular added copper decreased homocysteine (and thus, increased B12) [139]. Copper may positively influence the enterohepatic cycle and the flow of bile acids. Bile plays a part in normal absorption of vitamin B12 [144]. Presumably first pancreatic enzymes play a role in B12 adsorption, because they degrade R protein in the vitamin B12-R protein complex, followed by binding of vitamin B12 to intrinsic factor. Bile acids may dissociate the intrinsic factor-B12 complex at the receptor site in the ilium, enhancing the absorption of vitamin B12 [144]. Furthermore, vitamin B12 is stored in the liver and a large fraction exist in the enterohepatic circulation, using Rprotein and intrinsic factor, and treated exactly like the vitamin B12 from diet from the stomach.

Copper deficiency may be one of the contributions to vitamin B12 deficiency, but otherwise, B12 deficiency may be one of the contributions to copper deficiency. Homocysteine (more homocysteine results in less vitamin B12) decreased the protein levels of Cu chaperone COX17 (involved in intracellular copper transport), which was accompanied by a decrease in the activity of cytochrome c oxidase [135]. Thus B12 and copper are closely related.

A relation between vitamin B12, and thyroid hormone needs the enterohepatic circulation as explanation. Interesting in this context is the finding that administration of thyroid hormone (T3) leads to an increase of the maximal rate of bile acid synthesis (of hypothyroid rats showing diminished levels of bile acid synthesis [135b]. The fact that hypothyroidism correlates with a deficiency in vitamin D and B12, fits in the idea of the role of enterohepatic cycle in intestinal absorption of vitamin D and B12.

(3) Vitamin B12 and phase earth: Milt/Stomach meridian. Relation between blood emptiness and zinc.

Acupuncture (using acupuncture points to ‘feed’ Milt) may relax the stomach (by vagus nerve) and this will increase zinc and vitamin B12 absorption (optimizing intrinsic factor). Low gastric acid reduces zinc absorption [179] and the vagus nerve influences gastric acid production: in this way, stress decreases zinc absorption. In fact, also other nutrients depend on stomach functioning: Proton pump inhibitors are associated with an increased risk of vitamin and mineral deficiencies impacting vitamin b12, vitamin C, calcium iron and magnesium metabolism (hypomagnesia is combined with hypokalemia and hypocalcemia) [137 of 141].

Another association of B12 with phase earth is: Neuropathy in the legs are treated by acupuncture meridians located on the Stomach meridian, suggesting that vitamin B12 absorption in the stomach is an important contributing factor in this process. Chapter 6.2.6 describes the mutual influences between vitamin B12 and copper.

In summary, the rule in TCM that blood emptiness is a situation between yin emptiness and yang emptiness is decribed by vitamin B12, iron, copper and zinc and magnesium.

Internal Wind.

In acupuncture blood emptiness (copper deficiency, B12 deficiency) may become worse into a situation described as internal Wind. A characteristic of wind is the fast changing location and fluctuating intensity. Compared to transport of hormonal and chemical information, neural signaling is relative fast.
6.2.7 Pulse diagnosis and emptiness in yin and yang.

In acupuncture the pulse diagnosis is applied on the arteria radialis at the level of the processus styloidus, and other locations of arteries are also theoretically possible. This position on the left pulse indicates qualities of heart, liver, kidney (for instance) with respect to yin emptiness. On the right pulse it indicates qualities of pancreas, stomach and kidney (for instance) with respect to yang emptiness. We simplify to yin yang emptiness. Chapter 5.7 describes as source of copper: the release of ceruloplasmine by the liver, bounded with copper. Cells in the neighborhood of the liver first receives the copper. A cell located further away from the liver last receives the copper (if enough copper), and thus, yin emptiness is easier detected at the left pulse compared to the right pulse. Analog for zinc: the pancreas releases zinc during digestion. Probably a cell located further away from the pancreas receives the zinc ions at a later moment (if enough zinc). This is why yang emptiness easier is detected on the right pulse. This reasoning assumes that copper and zinc is transported, not only by the fast blood system, but also via interstitial fluid between cells and lymph. In acupuncture, the right pulse is called the yin pulse, and demonstrate yang emptiness. Vice versa, the left pulse is the yang pulse and demonstrate yin emptiness. Why? Copper en zinc compete with each other. In fact yin emptiness means a relative increase in yang, as dominating factor on the yang pulse. Yang emptiness means a relative increase in yin, as dominating factor on the yin pulse. The yin yang picture used by Traditional Chinese Medicine, within the black yin, a white dot, and within the white yang, a black dot is shown.

Another finding in acupuncture can also be explained according to the pulse diagnosis description as is used above.

After CVA (Wind Stroke): in first instance, left side of the body is a preferred location of blood emptiness (and blood stasis), and right side of the body is a preferred location of Qi emptiness (Fire and Flegm) [111]. Blood emptiness occurred by copper deficiency, and left is the most far location from the liver. Qi emptiness may be a weak form of zinc deficiency or yang emptiness.

6.2.8 Damp

Different kinds of damp exists. Damp is a complex term, TCM subdivide damp into dampcold and dampheat. For instance, chapter 5.5 (obesity) describes a relation between albumin and edema. Albumin is a main extracellular transporter of zinc, thus zinc and certain kinds of edema may be connected to zinc in first instance. Chapter 5.2 describes a zinc-dependent enzyme in the kidneys (NEP) involved in the back-resorption of sodium and water. Presumably, this zinc-dependent enzyme influences a relative retention of the urine, which contributes in the acupuncture term “damp”.

6.2.9 Phlegm and glycoproteins.

Natural occurring glycoproteins exist in the stomach, as mucus, with as main component glycoproteins. Glycoproteins often show a high viscosity. Other important functions of glycoproteins are certain hormones, antibodies, ceruloplasmine and proteins which affect folding of certain other proteins.

Too much glycoproteins, which means proteins bounded by too much glucose: these proteins are less efficient. Due to a bad functioning digestion, too much carbohydrates in the diet, will yield too much glucose molecules. Degenerative brain diseases like Alzheimer and Parkinson obtain unnatural glycosylated proteins (advanced glycation end products) in
the brain [98, 99].

Acupuncture describes the evolution of phlegm from damp. Points REN-13 and REN-14 in the neighborhood of the stomach and duodenum, and ST-40 on the stomach meridian are used after "Phlegm" diagnosis [1]. Phase Earth, Stomach and Pancreas (Spleen) is associated with zinc and functioning of insulin. "Phlegm in heart openings" used in acupuncture may be the situation as described in for instance Alzheimer’s disease. In acupuncture brain and heart are closely related. Phlegm is difficult to treat by acupuncture. Acupuncture advised an adaptation of the diet.

6.2.10 EPF

In acupuncture, the term external pathogen factor (EPF) is measured by a significant amount of tongue coating (and certain answers to diagnostic questions and certain pulse characteristics). Tongue coating consists of bacteria, large quantities of desquamated epithelial cells, blood metabolites, different kinds of food remnants and leucocytes, coming from periodontal pockets. Thin coating mainly consist of an increased keratinization, while increased tongue coating consists of bacteria or virus infection, which in traditional Chinese medicine is referred as EPF [113]. (Not associated with EPF is the positive correlation between the amount of tongue coating and consumed milk, yoghurt, cheese and pudding [113]).

Why can a wrong choice of acupuncture points exaggerate the virus infection? An answer may lie in the systematic cell danger response (CDR). Systematic cell danger response (CDR) is a defense against chemical, physical and biological threads, like virus infection, and heat, salt, pH, cadmium, or chronic psychic stress during childhood. In this defense mitochondria play a key role. During CDR the number of mitochondria is decreased. Goal of this adaptation is that: when cells with virus infection remain healthy, virus will replicate. Cells with a decrease in mitochondria are less healthy. Mitochondria senses virus by detecting diversion of electrons as NADH and NADPH, as a voltage drop, resulting in a decrease in electron flow in oxidative phosphorylation in mitochondria. If the stress is chronic, metabolic memory of past stress encounters is stored in the form of altered mitochondrial content (mitocellular hormesis) [114].

According our article, a decrease of number of mitochondria released intracellular copper. During circadian rhythm, these extra copper ions are presumably washed out by liver. It is known that mitochondria have a central role in copper homeostasis. Mitochondrial dysfunctions play a key role in copper imbalance [115]. Could an increase of copper and zinc stimulate the generation of mitochondria? In that case, cells become healthier to produce more viruses. This is related to the warning in acupuncture treatment to be careful, if an EPF is going on, because treatment by wrong chosen points let the imbalance become worse.

Furthermore, CDR also causes a decrease in vitamin D and B12, which activation processes takes place in mitochondria [114]. (thus, CDR decreases copper and the enterohepatic cycle?).

**EPF: the active phase of the immune system: role of liver protein synthesis.**
Fever is achieved by a prostaglandin-induced change in the relative rate of firing of the heat- and cold-sensitive neurons within the preoptic/anterior hypothalamus, the primary
temperature controlling region of the brain [116]. Following infection or an inflammatory response, muscle wasting occurs, by changes in protein metabolism [116]. The amino acids are used for synthesis of acute-phase proteins in liver, oxidized for energy, and expansion and secretion of immunoglobulins [116].

Immune response influences circulating levels of elevated glucocorticoids, glucagon insulin and growth hormone, and decreased thyroxin levels [116]. IL 1beta in the brain stimulate the release of corticotropin releasing factor and increases ACTH. IL1 inhibits the release of thyroxin stimulating hormone from the pituitary, and decreases circulating levels of T4 [116]. Hyperinsulinemia and hyperglucagononemia are common to most infection and inflammatory responses [116].

IL-1, IL-6 and TNF alpha, three cytokines of leukocytic origin, act in concrete to decrease food intake, increase resting energy expenditure, gluconeogenesis, glucose oxidation and hepatic synthesis of fatty acids and acute phase proteins, decrease fatty acids uptake by adipocytes and alter the distribution of zinc, iron and copper [116]. This is done by changing levels of circulating insulin, glucagon and corticosterone [116]. These changes occur in all tissue types.

Both copper and zinc are a modulator similar to calcium in cell signaling, and modulate membrane receptor function [117]. For instance in certain glutamate receptors [117] (see 8.0). For each of the ions, copper and zinc, there is an optimum concentration: both excess and limiting copper levels can modulate MAPKs with important consequences to cell fate [117]. Granulocytes take up pathogens by phagocytosis and production of reactive oxygen species (ROS), including the superoxide anion [118]. Superoxide anion is produced by NADPH oxidase, which is inhibited by zinc deficiency, as well as zinc excess [118].

During infection and inflammatory processes, a serum redistribution of iron, copper and zinc takes place [119]. The cytokines IL-1, TNFalfa and IL-6 are liberated by activated macrophages, in response to several stimuli, including exercise, trauma, stress, or infection [119]. (Cytokine is a hormone-like molecule, involved in cell-to-cell communication).

Iron: IL1 and TNFalfa induce the sequestration of iron mainly in liver, spleen and bone marrow tissues [119]. Thus, fe is sequestered in compartments which are nutritionally unavailable to bacteria and parasites [116].

Zinc: IL1 also induces the MT synthesis in the liver, which in turn binds the serum zinc [119]. By this process, zinc is replaced from serum to bind to MT in the liver (to protect the liver from ROS caused an increased synthesis of certain proteins) [119]. Also MT is increased by IL-1 in bone marrow and thymus., two essential organs for leukocyte development [118].

In vitro deficiencies of Zn, Cu and especially mg in the incubation medium impair release of IL-1 [116]. Supplementation of culture medium with Zn enhances the responses of IL-1 activated thymocytes [116].

Copper: IL-1,, IL-6 and TNF alpha induces in the liver an increased synthesis of acute-phase proteins like ceruloplasmin (serum transport of copper) and transferrin (transport of iron). The ceruloplasmin transports copper in the serum. Grubman: Copper complexes are needed in wound healing. Copper increases collagen synthesis and tissue repair. In addition the liver decreases synthesis of serum albumins (binding Zn in the serum)) [121]
The changed Cu/Zn in the plasma is needed in an acute stage of the immune system. Afterward acupuncture may reset the Cu/Zn ratio in the serum, to diminish the side effect of a changed Cu/Zn ratio.

In fact, it is the transport of Zn and copper which is needed. (according to [121] zinc should be added a period before vaccination and not during vaccination). A changed Cu/Zn in the plasma have side effects, which may be suitable for acupuncture to be addressed. Mostly, acupuncture is not efficient during acute infection (epf), because it may worsen the illness.

TCM advices to use acupuncture after the maximum crisis, for instance in the case of malaria [120]. In another example, in the case of hay fever, the acupuncture formula in the acute phase is totally different from the acupuncture afterward, in the autumn: dispersing versus feeding (collecting Cu and Zn around the acupuncture point?).

Zn supplementation in healthy subjects limit inflammatory cytokine production and oxidative stress during inflammation [118]. Zinc supplementation improves the response to vaccination, autoimmune diseases [118].

However, extremely high amounts of Zn simultaneous supplied together with administration of the vaccine were mostly ineffective or even counterproductive, whereas supplementation in advance of vaccination generally improves antibody formation [118]. In general: Parenteral zinc supplementation (direct in blood) aggravated the febrile response in patients undergoing an acute phase reaction [118].

7. Magnesium

Magnesium is an interesting ion, because deficiencies of magnesium leads to many symptoms, used in acupuncture diagnosis. Just as in acupuncture, life style is very important on magnesium homeostasis.

It is known, that zinc and magnesium inhibits each other, which the body may use in regulation processes. Both magnesium and calcium are bivalent, but magnesium has a greater electronegativity and thus exert greater pull on water molecules, compared to calcium. Thus, magnesium ions block calcium channels such as NMDA channels.

Interestingly, indirectly active zinc transport needs calcium [1b], and calcium is a concurrent of magnesium.

Because zinc play a central role in our article, magnesium should not be ignored. The magnesium-calcium concurrency can fit in the Wu Xing model by the distinction between phases Water Fire (magnesium) and phases Earth and Wood (calcium) (see chapter 6.1).

Publications exist on the subjects acupuncture and magnesium. Acupuncture may increase serum magnesium: after a number of acupuncture treatments of migraine patients, migraine disappeared in the majority of patients, and their serum magnesium (fasting blood sample in the morning) increased from the low value 0.58 mmol/L (normal: 0.8 to 1.2 mmol/L, in serum.) to 1.01 mmol/L in the normal range of healthy people [118b]. This is in agreement with the finding that migraine is characterized by a lower content of magnesium in the cerebrospinal fluid and in red blood cells, and the finding that
oral magnesium sulfate is effective for migraine [118b].

Mg and zinc compete with each other respect to phosphorylation: Phosphorylation of proteins by the protein tyrosine kinases (PTK) is antagonized by dephosphorylation by protein tyrosine phosphatase (PTP) [118]. The dephosphorylation by PTP can be limited through inhibition by zinc (among others) to keep the accent on the phosphorylated molecule [118].

Another example of phosphorylation is the effect of magnesium on protein kinase C which in turn effects zinc channels [147b, 147c].

[151b] At the cellular level, magnesium ions are highly compartmentalized within the cytoplasm, mitochondria, nucleus, and endoplasmic reticulum. [151b] Within these compartments magnesium is associated with phospholipids, chromatin, ATP, and other phosphonucleotides: total magnesium concentrations range between 15 and 8 mM within the cellular organelles, and between 4 to 5 mM in the cytoplasm.

Two of the phosphonucleotides to which magnesium likes to bind are the cofactors nicotinamide adenine dinucleotide (NADH) and nicotinamide adenine dinucleotide phosphate (NADPH): NADH added with a phosphate group. Despite the similarity in how proteins bind the two coenzymes, enzymes almost always show a high level of specificity for either NADH or NADPH. This specificity reflects the distinct metabolic roles of the respective coenzymes. A few exceptions to this general rule, and enzymes such as glucose-6-phosphate dehydrogenase, can use both coenzymes. NADH is important in catabolism, and in cellular respiration and oxidative phosphorylation, and in gluconeogenesis. Main function of NADPH is as a reducing agent in anabolism, with this coenzyme being involved in pathways such as fatty acid synthesis.

For instance, in a certain enzyme of certain bacteria: the magnesium-dependent NAD(P)(H)-binding domain in the nicotinoprotein methanol dehydrogenase with one zinc ion and two magnesium ions, and a tightly bound cofactor NAD(H) per subunit. [151c], The magnesium ions are essential for binding of cofactor NAD(P)(H).

(Interpreted in the hypotheses of yin of metals in the context of our article, magnesium may accelerates or decelerates both anabolic and catabolic processes together. This illustrates the complexity of Yin and Yang: Yin and Yang are relative terms: another yin yang couple is anabolic versus catabolic processes.)

First, different symptoms of magnesium deficiency will be related to acupuncture. How acupuncture’s local increase of zinc may increase magnesium effects on the other side of the body or meridian, by replacing zinc to the spot of needling, will be described in one of the next chapters.

7.1 lifestyle

Influence of lifestyle on magnesium is the following: A lifestyle intervention, certain exercises, enough sleep, and reducing total and saturated fat and increasing fiber intake, affects the intake magnesium, not copper and zinc [147]. With respect to food: [150] The most frequent source of Mg deficiency is an inadequate diet: magnesium is lost during the processing of food items. Phytate, fibre, alcohol, or an excess of phosphate and calcium attenuate the absorption of Mg. [149] Magnesium is involved in metabolism of fat, amino acids, and sugar. [150] Overeating (excess energy intake) is a risk factor for Mg deficit. [mg4]: High levels of fatty acid and protein may decrease uptake. [149] Mg plays a pivotal
role in insulin secretion from pancreatic beta cells. Because insulin by itself stimulates intracellular Mg uptake, Mg deficiency forms a vicious cycle, inhibiting insulin action. [148] Insulin may enhance cellular Mg uptake. [148] In the kidney it can decrease the excretion of Mg. With respect to exercise: [150] Moderate physical exercise as well as adequate rest and sleep under comfortable circumstances are important for the cells to accumulate intracellular minerals, including Mg. With respect to sleep: the minimal magnesium lost in urine occurs during the night (art). [mg4] Mineral losses in urine and sweat are more important. [149] Items increasing urinary Ca and Mg are: ethanol, glucose, fructose, casein, insulin, sodium, acid, heavy exercise, mental stress, cold exposure, over eating, Items that decrease urinary ca and Mg are: fasting, and low energy diet.

Traditional Chinese medicine emphasizes a certain life style: enough sleep, following the circadian rhythm, rest between activities, diet restrictions, that are related to an optimal condition of magnesium [151]. With respect to stress: adrenalin and cortisone, elevated during stress, increase the elimination of mg through the kidneys [151]. Mg reduces secretion of cortisol, hence lowering activity of the hypothalamus-pituitary axis [151]. With respect to enough sleep: Mg therapy is known to normalize night sleep and to restore the rem stage of dreaming [151]. With respect to diet: High consumption of protein, carbohydrates and fat foods increase the need for mg [151]. Extra food processing and refining leads to profound loss of many minerals including mg [151]. Reduced enteric adsorption of mg occurs with frequent consumption of coffee [151]. Alcohol, drugs and smoking considerably accelerate elimination of the mg from the body [151]. Food additives impede mg absorption [151]. Mg is used in the metabolism of diverse vitamin B’s [151]. With respect to environmental factors: Improper use of fertilizers augment mg deficiency in the cultural soil [151]. Temperature too high (living in a too warm climate) or too low is a factor causing deficit of mg, but also an intense rhythm of life, trauma, emotional stress, infection, heavy physical labor [151]. A small amount of mg is excreted with sweat [151]. A noticeable loss of mg people who regularly attend sauna, athletes [151]. Physical exercise appears to potentiate mg dependent enzymatic digestion of the excess of triglycerides. Mg deficiency contributes to the formation of the insulin resistance and glucose tolerance [151]. Some items of this list of lifestyle advices can be recognized in TCM food advices [151].

7.2 symptoms of magnesium deficiency

Symptoms due to magnesium deficiency, and similarity with signs used in acupuncture diagnosis are the following. For the most parts, the symptoms of mg deficiency are those reflecting an increased neuromuscular excitability coupled with vegetative dysfunction [151]. Reason for this relation with vegetative dysfunction can be that Mg is required in synthesis of cAMP (second messenger) [151]. Mg deficiency leads to a systematic reduction in activity of all varieties of the adenylate cyclases (second messengers): and consequently to quite a range of neurological effects [151]. Memory, olfactory responses [151]. Predispose to depression, suboptimal functioning of the thyroid gland, opioid receptor signaling, transduction from beta adrenergic receptors [151].

With respect to increased excitability, symptoms of mg deficiency are: shakes, twitching, knee jerk reflex, constantly moves [151]. With respect to vegetative dysfunction, symptoms are: feeling of lump in the throat (in fact a spasm of the pharynx), diarrhea, constipation, dizziness, headaches, increased fatigue (mental and physical), hypotension, faster breathing, hot flashes, pollakiuria (often urinating), lowered body temperature, feeling of cold, propensity to edema, nausea, vomiting [151].
With respect to behavior, symptoms of mg deficiency are: fear and anxiety, or apathy and depression [151]. Reduced ability to concentrate, somewhat impaired memory operation, persistent bad mood, irritability, nervousness, hyperactivity, additive behavior [151]. Other symptoms of mg deficiency are: pain in the back/the lumbar, muscle cramps, hypotonia, tingling in feed and palms, seizures, fragile nails, calcification of the mg deficiency tissues: gall bladder stones, kidney stones [151]. Mg is needed for a regular menstruation cycle, prevention of PMS, normal sleep, with respect to rem stage of dreaming [151]. Mg deficiency is characterized by a wide range of the bone pathologies including: osteoporosis, rheumatoid arthritis, gout, scoliosis [151]. Chronic mg deficiency is a predisposing factor for the formation of bronchial asthma and recurrent bronchitis [151]. Mg reduce spasm of smooth muscles surrounding bronchiole and reduce the release of histamine by the mast cells thus reducing inflammation [151]. Both mg and aspirin have equally powerful anticoagulant effect and inhibition of coagulation (in the process of thrombus formation) [151]. A magnesium deficiency leads to impaired immune response against true pathogens, and at the same time, to an increase in autoimmunity and inflammation [151].

7.2.2 Estrogen and magnesium:
[148:] During the menstrual cycle, a cyclic variation in Ca/Mg ratio was reported. There was a decrease in the level of ionized Mg at the time of ovulation, total Mg decreased only in the luteal phase. This indicates a role for estrogens and progesterone in Mg homeostasis. This may be due to the inhibitory effect of estrogen on PTH (parathyroid hormone) induces bone resorption, resulting in reduced plasma total Ca and stimulation of PTH secretion. (amount of estrogen is maximal during first half, after menstruation, before ovulation, (according to acupuncture: with an increase of yin), and progesterone during second half, (according to acupuncture with an increase of yang)) .

7.3 Biochemical transport of magnesium.

7.3.1 Mechanism of Mg influx (extracellular into intracellular).

Channel TRPM7 (member of the transient receptor potential channel family), with a specific permeation profile of Zn > Mg > Ca, transports magnesium into the cell. [149] Under standard physiological conditions, it is a primary Mg conducting ion channel that is regulated by intracellular concentration of Mg intern. [149] TRPM7 channel mediates Na and K efflux. [149] Another channel, MagT1 is a voltage-dependent magnesium transporter with channel-like properties. [149] MagT1 genes expression is high in kidney, colon, liver and heart, while weak in the intestine, brain, spleen and lung. [149] These MgT1 proteins creates currents that are induced by elevated Mg-extern but not by elevated concentrations of other divalent cat-ions.

7.3.2 Mg efflux (intracellular into extracellular).

[149] Extrusion of mg must occur against a steep electrochemical gradient. [149] Other driving forces: Na gradient must be used to transport the magnesium ion by secondary active transport mechanisms. [149] A Na/Mg exchanger is a candidate mechanism for the extrusion of mg. [149] This transporter is indirectly dependent on the Na/K pump, because the driving force for this exchange is the inwardly directed concentration gradient of Na+. 

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It is measured that the rate of Na/Mg antiport is dependent on mg-intern concentration. [148]: Extracellular Na is needed in the efflux of Mg. (This Na gradient is built with Na/K receptors, so K is also relevant in mg adsorption). [148]: This Na dependence may also be due to the activity of the Na K/ Cl symporter, that would provide Cl to a Mg/Cl symporter. [149] Alternatively, Na/Mg extrusion is activated by changes in intracellular Ca by a sodium-calcium exchanger.

**Channels of magnesium influx depend on zinc.**

Channels responsible for Mg influx show dependency of zinc: [149] Zinc is most efficient transported by channel TRPM7 of Mg influx, fitting in the idea that serum zinc may manipulate intracellular magnesium. [149] MagT1, a magnesium transporter with channel-like properties: its currents are inhibited by zinc.

### 7.3.3 vitamins D, B12 and copper, mg and zn.

As is described, copper may influence vitamin B12 and too much copper inhibits zinc. With respect to vitamin D: a decrease in the entero-hepatic cycle may decrease the fatty vitamin D.

Because many ions concentrations depend on each other, there is an optimum concentration of each, and adding one increased ion concentration in the diet may influence other ions. For instance a negative influence of increasing zinc may be caused by the resulting (presumably only intracellular) decrease of magnesium.

There exists also a relation between magnesium and vitamin D: magnesium regulates the production of 1,25OH2 D vitamin. Magnesium deficiency leads to reduced vitamin D in serum [59].

### 7.3.4 ATP, hormones and magnesium

[148]: The main factors affecting cytosolic free Mg are the concentration of nucleotides, especially ATP, and about two orders less for ADP. The cytosolic free Mg thus rise in cells in a poor energy state with less ATP. [148]: In the matrix free Mg, the metabolic state, inorganic phosphate and ATP reduce the free Mg, while a conversion of ATP to ADP increases it. The influx is stimulated by inorganic phosphate. In conclusion: it seems that the presence of magnesium is used as a measure of direct available energy (ATP molecules). Low magnesium means high energy state. It is not surprising that many hormones may influence Mg: [148]: Hormonal modulation of Mg: Parathyroid hormone PTH, calcitonin, vitamin D, insulin, glucagon, antidiuretic hormone, aldosterone and sex steroids have been reported to influence Mg balance. Alfa1 adrenergic agonists (phenyl epinephrine) and b adrenergic agonists, norepinephrine stimulate Mg efflux in cardiac and liver cells. Angiotensin II lower the cytosolic free Mg in vascular smooth muscle cells.

### 7.3.5 Two physiological systems that heavily rely on energy is regulation of vasodilatation and regulation of menstruation. Mechanism of influence of magnesium on hypertension.

An important aspect of magnesium with respect to interpretation of effect of acupuncture is the influence of magnesium on hypertension. [149] Hormones, like catecholamines, PGE2 and angiotensin II, activate the Na/Mg exchanger, to induce Mg extrusion. [149] Intracellular Mg decrease influences vasoconstriction after acute angiotensin II stimulation. Disturbance of the Na/Mg exchanger has been assumed to participate in the pathogenesis of primary hypertension. [149] Magnesium has an important role in the regulation of the metabolism of arachidonic acid. Depletion of intracellular Mg reduces exogenous
arachidonic acid incorporation into tissue phospholipids. Mg binds to protein kinase C, causes reduced activity. Protein kinase C mediates phosphorylation of enzymes that are involved in arachidonic acid incorporation. Sphingomyelinase is activated by Mg, vitamin D, TNF alpha, and IL-1. Sphingomyelinase plays an important regulatory role with a vaso-relaxant response in vascular smooth muscle.

7.3.6 Regulation of magnesium homeostasis.

[149] Plasma content in magnesium and calcium are controlled within narrow ranges and have no circadian variations. Urinary excretion of both magnesium and calcium have obvious circadian rhythm, higher in daytime and lower at night. [149] Thus, the serum Mg level may not reflect intracellular magnesium availability. [149] Magnesium homeostasis is preserved by control of intestinal absorption and losses through the urine as other minerals. [149] The excretion process from urine is the most powerful regulator for magnesium control mechanism. [148]: Key hormones that regulate the amounts of Ca, phosphate and Mg are PTH, vitamin D and calcitonin. Their actions are similar for Ca and Mg. These hormones exert their influence on Mg in the kidney, affecting Mg reabsorption (resulting in a minimal excretion of Mg during the night, this becomes a circadian rhythm of magnesium, with a maximum reabsorption during the night – according to acupuncture the period of maximum yin). PTH stimulates Mg reabsorptions, releases Mg from bone, and increases its absorption in the small intestine. Mg levels may influence PTH secretion through a feedback system. Chronic hypermagnesemia may suppress PTH secretion and cause disturbances in Ca homeostasis. [Mg4] Vitamin D enhances the intestinal absorption of Mg.

[149] In the case of Mg deficiency, Mg in the bone may be released into bloodstream. Vitamin D requires magnesium in its action. [149] Magnesium plays some roles in the regulation of calcium and bone metabolism. [149] For the minerals Na Ca Mg and p whose physiological pools include bone, are the signs and symptoms of deficit or excess poor, determination of the requirement and the upper limit is difficult. Calcium decreases Mg uptake. Optimal uptake of Mg requires sodium and phosphorous. [mg4]The physical signs of Mg deficiency are largely due to the associated hypocalcemia and hypokalemia.

The competition between magnesium and zinc is illustrated in the effect of magnesium on MT. Magnesium deficiency increased the mRNA levels of MT protein and zinc concentration in liver cells [158]. By intracellular decrease of zinc, magnesium will be active.

Mg and ATP.

Mg deficiency: Increased fatigue, mental and physical [151]. Mg is required in hydrolysis of ATP. 98% van mg in bones, skeletal muscle and soft tissues [151]. Tissues characterized by high metabolic activity and high concentration of mitochondria (namely brain (frontal cortex), heart, muscles, kidney, live rand placenta) [151]. The highest intracellular concentration of mg is found in mitochondria in the form of the mg-ATP complex [151].

Mg and catecholamines (like adrenaline).

Inverse correlation between the mg and the levels of catecholamines [151]. Mg is involved in the processes of degradation of catecholamines, acetylcholine, glycine [151]. Mg is
7.4 Interpretation of magnesium in terms of acupuncture and TCM. Magnesium was not mentioned by Yan after needling in an acupuncture point. Also, in the depression protocol of acupuncture, serum magnesium did not change [159]. Furthermore, the presence of intracellular magnesium determined its relation with for instance asthma, not the concentration of magnesium in the serum [156, 157]. Why is magnesium not changed around an acupuncture point and why still arm meridians (phase metal and fire) may influence a magnesium type function? Extraplacellular magnesium levels are under tight homeostatic regulation in the human body, and do not correlate well with total magnesium status or with the intracellular magnesium pool [160]. It seems, that this regulation of extracellular magnesium in the serum is more severe compared to zinc or copper. Reason may be that magnesium influences vessel diameter, influencing blood pressure (see section arm meridian phase fire).

In the context of the hypothetical circadian rhythm, magnesium may be increased in the following way. Acupuncture increases local zinc in the acupuncture point. This zinc may come from other places like intestines (for instance transported by cells of the immune system). Those other places notice a decrease in zinc. Acupuncture points of phase metal lie in the face and on the arms. Needling those points let zinc flow into those directions and increase there. Possible this zinc originates from the intestines, notices a decrease in zinc. After digestion, zinc is transported to diverse organs, among which the brain. May be magnesium fastens this decrease of zinc in the intestines. An important assumption is that after needling, zinc is transported along a distance and a decrease in zinc is felt on a location far away from the needled acupuncture point. This decrease in zinc is followed by an (only) local intracellular increase in magnesium. An argument of this way of manipulating magnesium by zinc is the following. Ca2+ and Zn2+ intracellular free concentrations, respectively [Ca2+]i and [Zn2+]i, are maintained at approximately 10-fold lower levels than the physiological extracellular concentration, thus generating a large chemical gradient for their mobilization into the cytosol [129]. By contrast, there is a much smaller difference (<twofold) between cytosolic free Mg2+ and extracellular free Mg2+ levels [129]. This effect on distance will be described in arm meridians, in Front Mu points, in the effect of distant points on a meridian, with an effect on the other side of the meridian, in a relation between Heart and Kidneys, the heart-kidney axis and yin emptiness in a broader context. Jing is addressed at DM-4 and REN-4 on lower back and abdomen respectively, with a local zinc increase, assuming to come from the head, yielding an increase in magnesium in the brain, to inhibit glutamate for instance.

7.4.1 Magnesium and Wei Qi

A competition exists between magnesium and zinc, and between magnesium and calcium. The competition between magnesium and calcium is indirectly illustrated in its need in phosphorylation. Magnesium is involved in enzymes that regulate phosphorylation (myosin kinases) and calcium is involved in enzymes that regulate dephosphorylation (myosin phosphatases). Reversible phosphorylation of proteins is an important regulatory mechanism. Phosphorylation results in a conformation change in the structure of the enzyme, causing them to become activated or deactivated [156]. In detail, a direct consequence of this phosphorylation can be applied to calcium transport: since magnesium is involved in calcium transport across the cellular membrane, (depending on phosphorylation) both phosphorylation and dephosphorylation are directly or indirectly influenced by intracellular magnesium levels. Furthermore, magnesium competes with
calcium for entry into cells through voltage-gated channels and receptors and inhibits calcium release from the sarcoplasmic reticulum [157]. (This relates magnesium deficiency to cramps).

7.4.2 Magnesium and meridians

Arm meridian phase Metal.

Wei Qi is associated with phase Metal. Wei Qi is defined as the efficiency of opening and closing the pores of the skin, related to warming of skin and muscles and protecting against cold [161]. A hypothetical connection between magnesium and Wei Qi is indicated by the correlation between a low concentration of magnesium and syndrome of Raynaud (m4: art lempert): Raynaud phenomenon is defined as an excessively reduced blood flow in response to cold or emotional stress, causing discoloration of the fingers, toes, and occasionally other areas. A reduced blood flow in the skin results in a situation that someone gets fast a cold sensation in a cold environment and fast a heat sensation in a warm environment. When there is not enough blood to transport temperature, one will easily sweat. Furthermore a reduced blood flow in the skin easily result in a dry skin. (Both transpiring and a dry skin are associated with yin emptiness). A direct influences of magnesium on phase metal is the following. Magnesium exerts a broncho-dilatory effect, probably by antagonizing calcium-mediated bronchoconstriction [162]. The other elements of Wu Xing are also influenced by yin emptiness (called kidney yin emptiness): Magnesium plays a role in activation of phagocytosis in pollinosis and is discussed in the literature in relation to acute rhinitis [163].

With respect to the immune system: Addition of magnesium ions in vitro, down regulate the expression of inflammatory mediators such as tumor necrosis factor alpha (TNF alpha) and nuclear factor k B (NFkB). Dysregulation of these and other inflammatory mediators has been linked to several inflammatory disorders, including asthma, arthritis, atherosclerosis and neuro inflammation [156, 171].

Presumably in skin diseases for instance, zinc and magnesium cooperate: hypothesis: there must be enough zinc, to feel an intracellular decrease in zinc, resulting in an increase of available magnesium.

The term “Po” (associated with phase metal) stands for physical consciousness. This may be reflected in MT. The number of MT may mirror the sensitivity of phase of wu xing, which is according to acupuncture theory also correlated with the sensitivity of the emotions associated with these phases of wu xing. Magnesium inhibit zinc and thus the intensity of emotions, obtaining an emotional distance. Falling sleep, phase water, needs a minimum of emotions, reflected in a minimum of MT. Indeed a deficiency of magnesium correlates with mood swings and magnesium shows similarity with lithium [164]. An association between magnesium and sleep is also made by the fact that magnesium is involved in synthesis of melatonin [165]. A relation between magnesium deficiency and tiredness is seen in the influence of magnesium in fibromyalgia [166]. In the context of “phase metal feeds phase water” Phase water: vitamin D is involved with the absorption of magnesium in the intestines [167].These items indicate a role of magnesium in the transition from metal to water.

Arm meridian phase fire.

Magnesium inhibits the glutamate receptor. A deficiency in magnesium can be associated
with insomnia, palpitations, and mood swings [154, 155]. Acupuncture influences BP by sympathetic nervous systems. Acupuncture on P-5, P-6, and LI-11 (all located on the arm) modulates activity of a location in the brain: the rostral ventrolateral medulla (rVLM). This area has cardiovascular premotor sympathetic neurons. Inhibition of rVLM results in large decreases in blood pressure. Enhanced sympathetic activity increases the secretion of renin and angiotensin [168]. Carotid sinus and aortic arch baroreceptors respond to changes in BP by modulating parasympathetic and sympathetic outflow (with a genetic component). Once this is chronic higher, the baroreflex resets to a higher blood pressure, with a higher sympathetic activity. Acupuncture may reset this chronic situation [168].

Magnesium supplements appeared to be protective against arrhythmias and death due to acute myocardial infarction of chronic heart failure, and suppresses the development of atherosclerosis [158]. Magnesium is absorbed via intestines. Different diseases of intestines show a magnesium deficiency. During digestion, there may be a wave in zinc concentration, followed by a decrease in zinc, which optimized magnesium absorption in the intestines. The organs associated with yang arm meridians are the small and large intestines.

7.4.3 Other phases related to magnesium:
Phase earth indirectly: diabetes patients and obese patients often show a deficiency in magnesium [154]. This may be caused by polyuria: with the urine also magnesium (and zinc) disappears. The founding that acupuncture increases the effect medicine (diuretics) against blood pressure [170] may be described by this deficiency in magnesium caused by polyuria. Interestingly Huang di nei jing described/found in Su Wen the syndrome Xiao ke, later interpreted as diabetic exhaustion [169]. This may fit with (or include) the idea of a combination of yin emptiness (magnesium) and yang emptiness (zinc).

Phase wood indirectly: Magnesium can be associated with cramps. This is used in acupuncture diagnosis to determine phase wood or blood emptiness in the liver (meridian). Hypothesis: during phase metal magnesium is optimal used, during phase wood, a relative minimum of magnesium is available but still magnesium is important. If there is a deficiency in magnesium, this may be felt in the phase (wood) that muscles of the limbs are most important to use. The word "indirectly" can be read as "not functionally by purpose by the autonomic nerves".

7.4.4 Frontmu points related to magnesium
Traditional Chinese medicine: There are twelve front-mu points, located on the chest or abdomen in close proximity to their respective corresponding organ (associated with the meridian) [1], but in most of the cases they are not located on their associated meridian. Still there is a distance between the acupuncture point and the organ. The mu points may be used to treat acute, excess or hot patterns of diseases, but are also used for deficient and chronic patterns of intestines and Stomach [deadman]. Frontmu and backshu points become tender in response to disharmony of their respective organ, which give them a diagnostic value. Because frontmu and backshu are (in most cases) not located on their associated meridian, needling does not influence channel disorders of their associated meridian.

The close proximity of the frontmu point to its corresponding organ yield a hint to its working mechanism. Needling increases zinc in the frontmu point. This zinc may be transported from the location of the organs. In an excess pattern the organs possesses enough zinc to deliver to the frontmu point. At the location of the organ, magnesium may
be increased (if enough magnesium is present, and it is not empty heat combined with yin emptiness, or magnesium emptiness).

On the other hand, the backshu points on the back are closely located to zinc sources and in general, backshu points are better points used in deficient cold patterns. Then there is a lesser zinc shift notices on other locations like the corresponding organ. In the case of emptiness needling of frontmu points may decrease zinc too much at the location of the organ, that side effects will occur (less zinc, then less MT, then relative too much copper).

7.4.5 Kidney heart axis

Traditional Chinese Medicine: The kidneys belong to phase water and the heart to phase fire. The kidneys and heart are said to mutually support each other, the kidney yin nourishing and moistening heart yin and restraining heart fire, and heart yang descending to warm the kidneys. Harmony between the kidneys and heart is one of the prerequisites for a stable and peaceful spirit. Disharmony may lead to agitation, insomnia, poor memory and fear, madness and rage.

Needling points on kidney meridian, KI-1, KI-3, relaxed stress of the heart. Zinc transports relative to the feet (decreasing yang emptiness on the feet), giving magnesium (yin is shown) in the body space to relaxes vessels. Points on the heart meridian will increases this effect on magnesium in the body.

7.4.6 Four gates.

The same reasoning may be given for another combination of acupuncture points, LI-4 and LIV-3, called “the four gates” is a strong receipt for increasing qi flow. This increase of qi has a characteristic of pain resolving. According to TCM, pain is defined as stagnation of energy flow. This obstruction may be chronic (acupuncture with a temporary relief) or temporal (simultaneous with the healing process in the body: acupuncture with a fastening healing aspect). Needling of these point on arm and leg increases (available) magnesium in the center of the body.

An acupuncture point located on the feet, BL-62 is used in mania, depression, and agitation. These illnesses depend on magnesium in the brain (magnesium shows similarity with lithium).

7.4.8 Eyes: influence of zinc on eye diseases and on the circadian rhythm (by the eyes), and influence of magnesium on eyes indirectly by condition of the blood vessels.

Many meridians start or end around the location of the eyes. Apparently the eyes play an important role in TCM physiology. Needling on acupuncture points on meridians in the neighborhood of the eyes causes a local increase of zinc [2]. Zinc has a beneficial effect on a number of eye diseases (cataracts, age-related macular degeneration, and even diabetic retinopathy, [180], [181]). Diverse enzymes of the physiology of the eyes have zinc ions as cofactor. In the context of our article: when in the evening, the eyes get tired. In the evening, relative more copper and less zinc ions are present in the blood (postulated in our article). To protect the eyes from the light they will close, which promotes to fall asleep.
Furthermore, zinc is a cofactor of superoxide dismutase and zinc induces MT synthesis. Both aspects have anti-oxidant properties.

Traditional Chinese medicine describes a close connection between hart meridian (vessels) and the eyes. The condition of blood vessels influences functioning of both the eyes and the heart [182]. Retina is strongly supported with blood: vulnerable blood vessels (atherosclerosis) have consequences for eyes (chance of eye diseases) and heart [182]. The occurrence of certain eye diseases like retinal arterial or retinal vein occlusions, cataracts, age-related macular degeneration, and increases in intraocular pressure may be of diagnostic value of heart diseases [182]. On the other hand, a treatment of eye disease is accompanied by a treatment of the condition of blood vessels. Magnesium ions promote the condition of blood vessels.

Dry eyes are associated with Yin emptiness and Liver meridian. One cause of dry eyes may be a bad condition of blood vessels on the eyes, although many other factors cause dry eyes, like quality of tears, number of eye winks, and medication [183]. Our article associates Yin emptiness with a deficiency of magnesium, and magnesium is related to condition of blood vessels. The distal acupuncture points on the feet: BL-62 and KID-6 may influence sleep by decreasing zinc and thus increasing magnesium in the rest of the body, to optimize blood vessel functioning.

7.4.9 Nails, magnesium and zinc.

Mineral zinc deficiency and iron deficiency can contribute to longitudinal ridges in the nail. Patients with soft flaky nails that are inclined to break or split may have significantly reduced plasma magnesium levels. Nail condition is associated with phase Wood emptiness [184].

7.4.10 Yin emptiness: from magnesium deficiency to copper deficiency and vitamin D deficiency

Different ions concentrations depend on each other: With respect to magnesium and copper: rats fed with a magnesium deficient diet get faster steatosis [129]. Assuming that steatosis correlates with a decrease in the entero hepatic cycle and this correlates with a smaller variation in copper in the body, this lead to the following hypothesis: (one of the types of) yin emptiness starts with magnesium emptiness and is followed by copper emptiness. It fits with our hypotheses, magnesium may contribute to the blood flow to the liver, to collect copper from the blood, to decrease copper during the night, in the circadian rhythm.

These issues about magnesium may be summarized to the following hypothesis: magnesium may show an intracellular circadian rhythm, according to phase metal (and fire), and may be influenced by increase of zinc in the arms, by muscle activity or by mimicking this by needling certain acupuncture points on the arm meridians. The autonomic nerve system may use zinc and copper, influencing thereby iron and magnesium, to optimize the circadian rhythm, influencing physiology and behavior. As cofactors these metal ions are very reactive and potent, and thus these molecules has to
be protected and concentrated on optimum times in day and night.

7.4.11 Magnesium and clinical application of the five shu-point and Xcleft points according to classical acupuncture theories.

Characteristics of the shu points are connected with their relative position distal/proximal on the arm and leg. Shu points are translated in terms of copper and zinc into terms of number of MT. Now we interpret them into terms of Points located in the area between the elbows and fingers and the knees and toes are among the most important points on the body, and have a wide application in threatening disorders of the head, chest, abdomen and back [1], and pain in meridians or organs. It is clearly demonstrated in the jing-well points, located near the nails of feet and hand. Jing well points are applied for clearing heat (excess, high fever, fullness, headache, read eyes, tinnitus, mania, insomnia, oppression and pain of the chest with dyspnoea) and restoring consciousness. Furthermore important (master) points of extra ordinary meridians are located on feet and hands.

Is this explained by a traveling of zinc to the other sides of the meridian, inducing an increase of magnesium at the other side? Magnesium is very important for brain function and widening arteries (by magnesium?) around the lung and heart decreases dyspnoea. Magnesium promotes vasodilatation of blood vessels. Applied to the most distal shu points: Jing-well: Jing-well points (near the nails) are applied for fullness below the heart and for clearing heat. Ying-spring points (on the root of the tow and finger) are applied for heat in the body. One of the blood functions is to distribute heat, and vasodilations promotes this effect. Less distal is the location of Xcleft points, used for acute conditions of pain (a stagnation of Qi or other substances) and of blood stagnation. This may also be translated in the vasodilation effect of magnesium. Luo points on fore arm and lower leg are used for emotional problems, caused by Qi stagnation (Hypomagnesia leads to hyper excitability of neurons, due to cellular calcium transport [148]. Mg reduces the activation of glutamatergic systems in the brain [mg4]). An exception of this trend are the source points, located on the inflection points of hand and feet, noticing the behavior related position of hand and feet. These source points influences the meridian as a whole coupled with the organ, coupled with the meridian. A second exception are the he sea points, located on elbow or knee. He sea points are applied to diarrhea, skin problems, and counterflow qi, which can be translated in zinc related properties of digestion. Again the projection of meridian into the map in the brain, because its relation to behavior and position of arm or leg in space counts.

7.4.12 Upper Burner, Middle Burner and Lower Burner, EPF, and magnesium, zinc and copper.

This chapter will first describe three locations of most optimal regulation of the ions, copper, zinc and magnesium. This optimal regulation is regarded as fitting of an optimal use of this ion in the context of a physiological state of behavior, like sleep, eating, or physical activity. UB will be associated with Cu, MB with Zn, and LW with Mg. Then the three burners: upper burner (UB), middle burner (MB) and lower burner (LB) are defined, and described in the context of the acupuncture term EPF. This is related to the actions of
the liver in the context of EPF, and hypotheses are formulated in this translation.

**Locations of optimal control of copper, zinc and magnesium respectively.**

This article emphasis the role of copper, zinc, magnesium in acupuncture. Acupuncture manipulates directly copper and zinc in the acupuncture points and in the serum. Magnesium, intracellular is only indirectly manipulated by the change in zinc serum, caused by the needle. Each of the ions are carefully regulated in the body. The main points of regulation of copper, zinc, and copper, driven by behavior, could be: (1) in the case of copper: initiation of physical activity. The decision to initiate activity lies in the brain, catecholamines, its synthesis is copper related. Catecholamines antagonize magnesium. In fact during sleep, the epiphysis secretes melatonin in the brain which bind copper, to collect copper, and send this to the liver (a hypothesis of this article). Ascending Yang.

In the case of zinc: initiation of eating: appetite is zinc related; the pancreas releases zinc by digestion enzymes. Yang emptiness.

In the case of magnesium: during sleep, less magnesium leaves the body by urinating. Cortisol and epinephrine (produced by glandulae adrenales) present during daytime increases the amount of magnesium that leaves the kidneys by urine. Mg inhibit the glutamate receptors in the brain during sleep. Yin emptiness.

These main points of regulation of copper, zinc and magnesium are at a distance of each other (kidney, pancreas, brain), to prevent that the measured change of ions is determined by competition by another ion (regulation point), by concurrence between zinc and magnesium, or concurrence between zinc and copper). The three burners are defined as the functioning of a group of organs: UB primarily includes Lung and pericardium, MB primarily includes stomach and spleen, and LB primarily includes kidney and liver [145]. We postulate the hypotheses that LB, MB and UB are regulation points of optimal control of mg, zn and cu respectively.

What does this hypothesis mean in the context of the regular main role of the liver during EPF? During EPF, in the case of infection, in reaction to cytokines, the liver produces: (1) more ceruloplasmin, the transport protein of copper in the serum, to obtain an increase of copper in serum, and (2) less albumins, the transport protein of zinc in the serum, to obtain an decrease of zinc in the serum, to receive more zinc in the liver.

Acupuncture: Rule of transmission of EPF in the case of Warm Heat diseases ([154] pp 248): Warm heat diseases begin in the upper burner. If upper burner disease is not resolved, this results in transmission to the middle burner spleen and stomach and, if the middle burner disease is not resolved, this results in transmission to the lower burner liver and kidney. Then, the following assumption is made: the main role of the liver in EPF could be translated in time aspects. These are: first process (1), increase of copper in serum, second process (2), decrease of zinc in serum. When the disease is most severe, zinc in the serum is such low, that it cannot manipulate magnesium, process (3). Acupuncture defines the burners in terms of effective acupuncture points to lighten the symptoms of the changed ion in the particular process. This fits in the hypothetical definition of the three burners, namely located on a point of maximal physiological
manipulation of the ion. The sequence of affected Burners can be recognized in the sequence of the three processes: first EPF in UB, then in MB, then in LB.

In our definition of UB, UB also consists of heart and brain (in TCM in the definition of heart, brain is included). Although brain is protected for EPF by the blood brain barrier, it is a source of manipulation of copper. Copper may be regulated by acupuncture on DM20 and DM16 influencing the current of the cerebro-spinal fluid. In acupuncture, cautions should be made in acute EPF: treatment is advises only at certain stages of the disease.

Overwork, Yin emptiness and magnesium deficiency.

Chronic overwork is related to yin emptiness, according to TCM. Our article relates yin with magnesium. [151] found a relation between magnesium deficiency and an increased cortisol level (starting with magnesium deficiency).

Hypomagnesaemia has been observed in association with metabolic syndrome [151b]. Rats exposed to a magnesium deficient diet, presented decreased glucose accumulation into the hepatocytes and an increased conversion of cortisone to cortisol (by enhanced glucose 9 phosphate transport into ER - caused by an enhanced hydrolysis by the glucose6phosphatase and conversion to 6phosphogluconolactone by the glucose6phosphate dehydrogenase. The latter process resulted in the increased generation of NADPH within the ER and the increased conversion of cortisone to cortisol by the 11bhydroxysteroid dehydrogenase type 1) [151b]. This stimulated cortisol production (perhaps for gluconeogenetic purposes) sets the condition for hepatic insulin resistance [151b].

8.0 Qi stagnation

Glutamate, the NMDA glutamate receptor and magnesium, zinc and copper

Pools of zinc and/or copper are stored in synaptic vesicles and the terminals of some, mostly glutamatergic neurons [152c]. The NMDA receptor is a glutamate receptor. It is activated when glutamate and glycine bind to it. And when activated, it allows (nonselective) sodium ions and small amounts of calcium ions into the cell, and potassium ions out of a cell (by their own channels). The NMDA channel is blocked by extracellular magnesium, zinc and copper:

(1) Magnesium: The NMDA receptor is blocked by magnesium at resting membrane potential. To unblock the channel, the postsynaptic cell must be depolarized. The
channel is open when simultaneously glutamate is bound to the receptor and the postsynaptic cell is depolarized (which removes the magnesium blocking the channel).

(2) Zinc: Zinc will strongly inhibit NMDA receptor-mediated charge transfer only when zinc is present in the cleft before stimulation [152b]. This may represent a mechanism for synaptic short-term depression of the NMDA receptor response [152b].

(3) Copper and zinc: independent? Zinc - and copper ions generally block NMDA current activity in a noncompetitive and voltage independent manner (wiki). Zinc may potentiate or inhibit the current depending on neural activity. In a number of cases copper and zinc show an opposite effect with respect to each other: With respect to the GluN2A (glutamate) receptor: copper caused a potentiation of the subsequent intracellular calcium response, while the same dose of zinc alone was ineffective (for calcium) [30]. When the two metals copper and zinc, were applied together, they did not produce any effect in the response immediately following, but the potentiating effect of copper was restored after washout of zinc [30]. As the GluN2A receptor is subjected to high affinity tonic block by zinc, it is possible that copper potentiates that GluNR channel by removing this inhibition [30]. Copper potentiation was prevented in the presence of excess zinc [30]. This interplay is reminiscent of that reported between copper and zinc in GABA receptor channels [30].

(4) Copper: while zinc only reduce glutamate-mediated excitatory transmission, copper completely eliminated all excitatory activity [152c]. Copper has a higher affinity than zinc for most substrates, thus affinity differences between zinc and copper could contribute to differences in their effects [152c]. According to the hypotheses in our article, Qi stagnation may merely be regarded as a kind of minimal copper.

The NMDA receptor is modulated by a number of endogenous and exogenous compounds (for instance glutathione) and play a key role in physiological (memory function) and pathological processes. Abnormal expression of certain glutamate receptors may play a role in schizophrenia and in chronic neurodegenerative diseases [30].

We will compare the influence of magnesium, copper and zinc on the glutamate NMDA receptor with a term in TCM, Qi stagnation (8.0.1). Then we describe a role of copper, zinc and magnesium in the memory task of NMDA receptors (8.0.2).

8.0.1 Glutamate and pain, depression: Qi stagnation.

Patients with severe chronic pain in an extremity, developed after trauma or surgery, with dystrophic signs and symptoms, show increased plasma levels of glutamate [152d]. Plasma glutamate is elevated in patients with migraine. If migraine was combined with fibromyalgia glutamate levels were higher. Plasma levels of glutamate were reported to be increased in patients with depression [152d].

Zinc, magnesium and copper have direct anti nociceptive effects [152e]. These data are
interpreted to mean anti nociceptive effect in central and visceral pain tests [152e]. This fits with the idea that zinc, magnesium and copper perform an inhibitory role on the NMDA receptor.

A medicine against pain palmitoylethanolamide (PEA) may be related to Qi stagnation. PEA is an endogenous fatty acid amide displays neuroprotective actions, on glutamate release from rat [152f]. Therefore, a reduction in glutamate release may be a promising neuroprotective strategy [152f]. Because the excitotoxicity caused by excessive glutamate release is a critical element in the pathogenesis of acute and chronic brain disorders, the ability of PEA to depress glutamate release may be one of the mechanism underlying neuroprotection [152f].

8.0.2 Glutamate NMDA receptor and memory.

An important characteristic of the NMDA receptor is its involvement in synaptic plasticity. Memories is thought to be encoded by modification of synaptic strength, and learning is based on the ability to change the strength of chemical synapses, based on recent patterns activity. The NMDA receptors (in the hippocampus) are involved in learning. Assume, learning process of the strength of chemical synapses consists of: first there is activity, then there is a process to strengthen the synapse, during an inactive state. Translated in this magnesium zinc and copper story, this could hypothetically fit as, first activation of glutamate, then inhibition by the zinc which was released together with the glutamate. Zinc is for short term memory on the NMDA receptor (yang). This process of update of synapse strength may be prolonged during the night, when magnesium is bound on the NMDA receptor (yin: slower process than yang; optimum of magnesium occurs during the night; depolarization removes magnesium from the NMDA receptor, not zinc, which binding is independent of depolarization). So it could be possible that magnesium, zinc and may be copper may generate (or follow) a kind of context, in which the synapse is biochemically strengthened. According to the assumed circadian rhythm in zinc and copper, copper binding to the NMDA receptor may be a compensation for the circadian decrease in zinc during the day, to keep glutamate receptor during the day in a kind of homeostasis.

8.1 Polyuria, Yin and Yang.

Polyuria depletes diverse minerals. A side effect of diuretic therapy is increased urinary loss of K and Mg ([171], [172]). This explains why kidney-yang emptiness based on polyuria causes both kidney yang emptiness (decrease in zinc) and kidney yin emptiness (decrease in magnesium). Polyuria is for instance a symptom in diabetes mellitus. (Same reasoning for sweat occurring during physical activity (if severe: hyperhidrosis) causes a decrease in magnesium and zinc [173].) The term "steaming bone disorder" used in traditional Chinese medicine in the case of severe hyperhidrosis during the night, may refer to the loss of minerals: bones contain a storage of minerals, like magnesium, zinc and calcium.
8.2 Moxibustion

Moxibustion is a TCM therapy using moxa made from dried mugwort (Artemisia argyi) pressed in a cigar shaped stick. This stick is burnt in the neighbourhood of the skin. Practitioners use moxa to warm regions and meridian points with the intention of stimulating circulation through the points and inducing a smoother flow of energy. Moxibustion at RM-8 on the belly increases zinc and zinc/copper ratio in the serum [143]. According to our article, zinc deficiency is related to yang emptiness, which fits with the use of moxibustion in the case of yang emptiness.

De novo lipogenesis and zinc.

An increased de novo lipogenesis needs zinc. Diverse glands (f.i. the prostate) uses de novo lipogenesis. Mammary glands de novo lipogenesis contributes significantly to milk fat [176]. But also growing tumors, and certain viral infections uses de novo lipogenesis [177]. This may be related to the acupuncture rule to prevent treatment in the case of growing cancer and in the case of an acute infection (EPF).
### 8.3 Summary of TCM terms regular expressed

Table 1 shows ion deficiencies in different TCM conditions: Yin emptiness, Blood emptiness, Damp and Yang emptiness. Interestingly, in 2009, Yan measured Fe, Cu and Zn changes in needled acupuncture points. Thus, at the level of the neighborhood of the needled acupuncture point, locally emptiness in Blood, Yin or Yang, or will at least temporary decrease.

<table>
<thead>
<tr>
<th>TCM term</th>
<th>Ion deficiency</th>
<th>TCM term</th>
<th>Ion deficiency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood emptiness</td>
<td>Fe↓, Cu↓, (e.g.) temporary blood loss (including loss of Fe, en Cu)</td>
<td>Damp</td>
<td>Zn↓, Cu↓ (damp cold: Zn&gt;Cu; damp heat: Cu&gt;Zn)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Phlegm</td>
<td>Entero hepatic cycle (Cu)↓</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Jing emptiness</td>
<td>physiological unnatural glycoproteins</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Qi stagnation</td>
<td></td>
</tr>
<tr>
<td>Yin emptiness</td>
<td>Fe↓ , Cu↓ and/or Mg ↓ (e.g.) Vit.B12↓</td>
<td>Yang emptiness</td>
<td>Zn↓</td>
</tr>
<tr>
<td></td>
<td>Chronic moisture deficiency</td>
<td></td>
<td>Glucose to cell ↓</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>e.g. many times urinating</td>
</tr>
<tr>
<td>Yang</td>
<td>Cu (ATP, dopamine)</td>
<td>Yin</td>
<td>Zn (glucose, steroid hormones, lipids)</td>
</tr>
</tbody>
</table>

**Table 1**: hypothetical relations between TCM term and some ion deficiencies. Heat, cold and other local stagnations are not included in the table. They can occur in all the TCM systems (meridians). These stagnations are a lack of flexibility and lack of adaptation of changes in energy needs during the circadian rhythm. All terms in the table may lead to extra stagnations in the form of for instance Heat, or Cold.

### 9 DISCUSSION

**Central role of metallothionein**

Central in the model of acupuncture described in this article is the protein metallothionein.
(MT). Because there are relatively many factors which influence MT, cytokines, stress hormones, catecholamines, glucocorticoids, nitric oxide production inhibitors, research on MT in vivo is difficult [16]. Thus the main role of this important protein MT is not yet defined in the literature [22].

This article uses characteristics of MT, the transport protein of copper and zinc ions, to make a sketch of a work mechanism of acupuncture. (MT does not bind iron ions.) Copper and zinc compete to bind on MT. Copper has a stronger binding energy on MT than zinc. But zinc increases the synthesis of MT, by increasing messenger RNA of MT. This can be related to the regulation of measures of stiffness’s of acupuncture points, a central aspect in the work mechanism of acupuncture.

Can copper also be responsible for a direct increase of MT synthesis? Nontoxic copper levels do not induce MT, although it is often bound to MT in vivo [16, 153, 174]. Cu induced expression of MT gene is likely to arise from secondary effects, such as the cu-induced changes in zinc pools [174].

Another indication that copper does not induce MT synthesis is the measurement that copper does not bind to MTF-1. The induction of MT by zinc occurs by binding of zinc to metal transcription factor (MTF-1). This protein bind to metal response elements (MREs) in the premotor region, which in turn initiates MT gene transcription. MTF-1 knockout mice die in utero from liver failure [16]. So zinc and not copper increases the synthesis of MT.

**Hypotheses.**

To obtain a valid work mechanism of acupuncture, the following hypotheses have to be tested, according to the model of our article: (1) copper and zinc play a regulating role in the mechanism of acupuncture. (2) A circadian rhythm exists in serum zinc and indirectly in (liver?) copper. (3) The increase of zinc in the needled acupuncture point stimulates locally the expression of metallothionein. (4) The local ratio of copper and zinc ions on MT determines the stiffness locally on the acupuncture point. (5) In the brain, a map of acupuncture points and meridians exist. (6) After needling, it is supposed that new MT is produced. (7) Metabolic syndrome is related to a decrease in EHC and a relative low MT concentration. (8) In the circadian rhythm of copper and zinc, liver (via EHC) and pineal gland (via melatonin) play a regulating role. (9) Different concepts used in acupuncture are expressed in terms of copper and zinc: damp (MT low), yin (zinc), yang (copper), yin emptiness (copper deficiency, magnesium deficiency), yang emptiness (zinc deficiency), blood emptiness (copper and iron deficiency), jing (severe copper and zinc deficiency, magnesium deficiency), and back shu points (zinc). (10) The ratio of copper and zinc ions on MT also follows the phase of Wu Xing and thus its emotion and behavior. (11) Shu transporting points on a meridian may partly be explained by their relative number of mRNA of MT, i.e. relative between the different shu transporting points on the same meridian. (12) Vitamin D deficiency is related with Jing and caused by a low functioning EHC and kidneys. (13) A role of vitamin B12 in blood emptiness. (14) A role of number of mitochondria in reaction of damage of the cell. (15) After needling, zinc is transported along a distance and a decrease in zinc is felt on a location far away from the needled acupuncture point. This decrease in zinc is followed by an (only) local intracellular increase in magnesium. (16) Is it physiologically possible to transport the small copper and zinc ions via interstitial fluid and via direct cell-cell interaction? With respect to cell-cell interaction: gap junctions between certain cells allow small molecules to diffuse between adjacent cells through which ions, sugar and other small molecules can pass.

Another interesting question is, if acupuncture changes the number of mitochondria in certain cells. Does a changed number of mitochondria in a location in an organ result in
changed local stiffness in the local connective tissue surrounding this organ? With respect to magnesium: if its maximum during circadian rhythm is during sleep, a period of restauration, yin building: are phosphorylation processes (for instance of the zinc potassium channel [147b], [147c]) optimal during the night?

**Other models of acupuncture.**

Other existing models of acupuncture use concepts of meridians, direct measureable in the periphery. Current models of acupuncture suggest a convergence of a neurophysiology model, a connective tissue model and a growth control model [175]. In the model of [175] interstitial connective tissue locations are points of a kind of bio electric potential field in which direction diverse molecules may migrate.

Our article may fit in the neurophysiology part, and includes a role of the highly reactive copper - and zinc ions, in their biological role as cofactors in enzymes. Their role as cofactor explains some influences of acupuncture on diverse organs and on connective tissue. Information of the acupuncture points is read by the neural system, and at the level of the brain associations are made with movements and behavior, to generate a meridian concept. This is a process of pattern recognition and neural networks. Another example of a neural network in the brain is [178]. Neural networks learn by association of input-output patterns, eventually irregular patterns, and store the learned information in their synapses. In our model, no flow of molecules along meridians is needed.

Another contribution in the physiological model of working mechanism of acupuncture is the hormone endorphin, with (among others) a anti nociceptive effect, and show down regulation properties in inflammatory processes [178b]. A link with cofactors zinc, copper or magnesium is possible but not jet found, this is why it is mentioned only in the discussion.

**Supplementation of zinc in the diet?**

One should be cautious with supplementation of zinc, because high intracellular zinc promote apoptosis. First it should be demonstrated that a zinc deficiency has occurred. To determine if deficiency in zinc is present, serum zinc and lymphocyte zinc should be measured [1b]. It is important that symptoms of zinc deficiency are visible, and are in remission after supplementation of zinc [1b].

Practically 98% of the zinc is located in intracellular space [1b]. In contrast, the concentration of free zinc inside the cell is lower than in extracellular space (and blood serum) [1b]. In fact healthiness is determined by a proper membrane function [1b]. Influx in the cell of the free zinc is a passive process due to the electrochemical gradient [1b]. Transport of zinc between organelles and cytoplasm inside the cell , and efflux of zinc is (sometimes also passive but mainly) an active process [1b]. Intracellular a potassium-zinc anti-transport (and chloride-bicarbonate anti-transport) takes place [1b]. The efflux includes a calcium zinc anti-transport channel [1b]. A proper membrane function is important, thus potassium and calcium are important in the context of zinc transport. Another example of (dis?) balance between ions is: high serum zinc is accompanied by deficiencies of calcium, copper, iron and by anemia. Combined zinc-magnesium-calcium is available (calcium and magnesium inhibit each other if unbalanced). In fact, multivitamin for elderly contain a bigger fraction of zinc,
magnesium and calcium. An advantage of natural products is their balanced ions. Hopefully, this article gives the impression that different ions are important and different balances between ions act in accompany, guiding or following important physiological processes. As will be described, acupuncture may temporarily relocate zinc, to stimulate certain physiological processes.

Another aspect of supplementation of zinc in diet, that this may decrease copper: zinc induces intestinal MT, which sequesters copper in the mucosal cell and prevents its transfer into the circulation. Because intestinal cells turnover is approximately every 6 days, then MT bound copper is removed in the stool [16].

Furthermore, zinc salts inhibit gastric acid secretion [179], and low gastric acid reduces zinc absorption.

According to the rough scheme of our article: if MT is a (abstract) measure of phase of copper/zinc ratio in the circadian rhythm, than adding too much zinc in the diet may disturb this coding. It may be different in the case of a zinc deficiency. Other research [22] shows that a physiological increase of zinc ions, (occurring in physiological processes) increases MT in hepatocytes. Hepatocytes from MT/-/- mice exposed to physiological zinc concentrations show little effect in glycolysis. In contrast, hepatocytes from MT/-/- mice exposed to physiological zinc concentrations, show a dose dependent relation between increased intracellular zinc, hepatic MT and glycolysis [16].

Furthermore magnesium and zinc absorption inhibits each other, and also: calcium and iron inhibits zinc absorption. With regards to intestinal absorption of copper and iron ion already display a competitive inhibition [126, 128].

Chapter 4.1.1 describes the influence of zinc on glutamate receptors, the influence of copper on glutamate receptors, but the inertness of copper and zinc together, with respect to the glutamate receptors [21]. Furthermore, the concentration of copper matters: a low performs total different effect on the receptors than a higher concentration of copper. Thus, there exist a harmony between a circadian variation in the ratio between copper and zinc. Just adding copper and zinc together will possibly not reach the brain directly.

Dietary Zn2+ deficiency decreases T cell activation, shifts TH1 responses toward TH2 responses, reduces the cytotoxic function of NK and NKT cells, and impairs cytokine production by mast cells and NK cells [129]. Zn2+ supplementation restores these functions but also induces chronic inflammatory responses and suppresses TH17 development in the elderly [129].(is magnesium a factor in this last process? In rodents, Mg2+ deficiency induced by low Mg2+ diet leads to decreased antibody production, thymic involution, and chronic inflammation [129]).

Different ions concentrations depend on each other: As is earlier described, magnesium influences steatosis [129], which may be correlated with variation in copper and which may decrease vitamin D absorption by EHC cycle.

Magnesium and copper: Rats fed with a magnesium deficient diet get faster steatosis [129]. Assuming that steatosis correlates with a decrease in the entero hepatic cycle and this correlates with a smaller variation in copper in the body, this lead to the following hypothesis: (one of the types of) yin emptiness starts with magnesium emptiness and is followed by copper emptiness. As is described, copper may influence vitamin B12.

Magnesium and zinc. In fact, if magnesium is stimulated by an decrease in zinc (serum) then for efficient working of magnesium there must be enough zinc. The idea of working
of changes in zinc concentration is written in [130].

Another way to increase minerals (zinc, copper, iron, magnesium, etc) in the serum is by exercises, which is measured by Bicer (2011) [130]. In fact if there is a deficiency in minerals like zinc, magnesium, then it is preferred to add these minerals in the diet or by a multivitamin. Acupuncture changes temporary the distribution of minerals, which first has to be present.

Diverse chronic diseases show a vitamin 25OH-D deficiency. When vitamin 25OH-D3 was administrated at 3 weeks of age before the onset of insulinitis, it effectively prevented the progression of diabetes in non-obese diabetic mice. In intestinal diseases, like coeliac disease [95, 96] a decrease is measured in zinc and copper iron, vitamin B12 and vitamin D.

10 CONCLUSION

This article describes how processes are dependent on copper and zinc and how these processes can be fit in acupuncture theory. Starting point is the measurement that needling an acupuncture point stimulates a local release of zinc ions. Using the properties of metallothionein, MT, the local effect of needling an acupuncture point can be described by a local change of softness (emptiness) or stiffness (repletion) into normal elasticity of the connective tissue. Acupuncture may change or reset the ratio of copper and zinc ions (on MT) into normal proportions.

The brain receives by proprioception an overall picture of the amounts of emptiness and repletion of all acupuncture points. This picture or neural map represents in fact a distribution of copper ions over the surface of the body, represented by acupuncture points. At this level, meridians are constructed. Associations are made between this map and sensory systems, phase of Wu Xing, hormones, and neurotransmitters. In response to the activity pattern on the map, the brain will plan a next physiological activity, in other words a - by habit - associated useful second behavior. This response can be for instance a release of hormones and neurotransmitters, or a change in respiration and blood pressure. Acupuncture may exert its working by manipulating this response a little. In particular behavior, some organs are active other not. The vagus nerve convey mechanical, chemical, osmotic thermal and possibly noicceptive information of organs to the brain. Mechanical is also measured: changes in stiffness of connective tissue by the copper dependent lysyloxidase. A local damage of an organ (a virus, heat of other aspects) causes cell defense response in which the number of mitochondria decreases. Then the damage cell is less active and viruses are not easily produced. Acetylcholine may counteract this. The vagus nerve projects from brain to organs: via the neurotransmitter acetylcholine. Probably the damaged location on the organ receives less acetylcholine, to permit the cell defense response. This projected map (with the local deviation representing the local damage on a particular organ) in the brain is possibly also projected to tongue, pulse and other parts like feet and hand.

Copper and zinc ions concentration in an acupuncture point, determining its stiffness, are influenced by: (1) five types of movements expressed as the five phases of the Wu Xing cycle used in acupuncture, and associated with five types of behavior (2) local body temperature, (3) diseases like headache, depression and metabolic syndrome, (4) some
regular medicine, (5) menstruation cycle, (6) circadian rhythm, and (7) diet. Copper and zinc both influences energy: both ions regulate ATP production in each cell, copper by increase, and zinc by inhibition. During inhibition by zinc, de novo lipogenesis takes place in certain cells, like in the production of hormones in certain glands or surfactant in alveoli cells.

Different acupuncture terms can be expressed in terms of copper, zinc and iron: damp (MT low), yin emptiness (copper and magnesium deficiency), yang emptiness (zinc deficiency), blood emptiness (copper and iron deficiency), jing (severe copper and zinc and magnesium deficiency). (9) The ratio of copper and zinc ions on MT also follows the phase of Wu Xing and thus emotion and behavior correlating with these phases. (10) Shu transporting points by their relative number of mRNA of MT. (11) Vitamin B12 and vitamin D deficiencies possibly influences acupuncture diagnostics with respect to yin emptiness and it may be interesting to test if yin emptiness (of kidney and liver) is correlated with deficiency in D and B12. Are these deficiencies more severe if rising liver yang is included? Does adding these vitamins influence (in case of measured deficiency) the result of acupuncture treatment. Does B12 deficiency focus on hartyin emptiness and D deficiency on kidney and liver yin emptiness? This could be the case, if The biggest influence of vitamin B12 is on the brain (upper part of the body) and vitamin D act on the whole body.

Acupuncture manipulates directly copper and zinc in the acupuncture points and in the serum. Magnesium, intracellular is only indirectly manipulated by the change in zinc serum, caused by the needle. This idea needs further research.

This description explains why acupuncture differs between two persons. It is only equal if these persons show the same copper and zinc pattern, in other words, same main complaint, behavior, circumstances, genetics, and same ancillary complaints.

The ratio of copper and zinc may follow the functioning of the immune system, and the de novo lipogenesis and cholesterol synthesis in for instance alveolic cells. Zinc ions inhibit ATP production. Copper dominates during daytime, which is the time of physical work. The circadian rhythm is regulated by the liver (timed by digestion) and the pineal gland (timed by light). During the sleep period, melatonin catches copper ions and the liver takes copper ions from the blood. During breakfast zinc ions are received again by the intake of food and the release of digestive proteins. In the next stage, start of physical work, zinc declines, because copper increases caused by an increase of epinephrine. Then, more copper ions will be available for the muscles to fasten ATP production, and to regulate dopamine.

Finally, effects of copper, zinc and/or magnesium are associated with the “Three Burners: Upper Burner, Middle Burner and Lower Burner” respectively in the case of copper: initiation of physical activity. The decision to initiate activity lies in the brain, catecholamines, its synthesis is copper related. Catecholamines antagonize magnesium. In fact during sleep, the epiphysis secretes melatonin in the brain which bind copper, to collect copper, and send this to the liver (a hypothesis of this article). In the case of zinc: initiation of eating: appetite is zinc related; the pancreas releases zinc by digestion enzymes. In the case of magnesium: during sleep, less magnesium leaves the body by urinating. Cortisol and epinephrine (produced by glandulae adrenales) present during daytime increases the amount of magnesium that leaves the kidneys by urine. These main points of regulation of copper, zinc and magnesium are at a distance of each other (kidney, pancreas, brain), to prevent that the measured change of ions is determined by another ion (regulation point). by concurrence between zinc and magnesium, or
concurrence between zinc and copper).

It should be kept in mind that a complaint or symptom have potentially many causes, depending on other characteristics and symptoms, not only on copper, magnesium, and zinc. If (other) zinc, magnesium, or copper related symptoms are also present, as protocols of diagnosis in acupuncture demands, acupuncture is a possible try. Research testing the validity of acupuncture for a certain complaint should only be applied to patients with the main complaint combined with zinc, magnesium, or copper related symptoms, recognized as a pattern in acupuncture diagnosis. Furthermore, the acupuncture treatment should decrease the symptoms used in the acupuncture diagnosis.

In fact, acupuncture may optimize these circadian variations in liver copper and serum zinc ions. A temporary locally increase in copper and zinc may result in a temporary push of the self-healing capacity. Clearly, inflammation regulates/shows a big influence on copper and zinc concentration. Acupuncture uses little inflammations on defined locations on the body to exert its effect.

11 LITERATURE


[76] Song CH, Kim YH, Jung KI (2012). Association of zinc and copper in serum and hair with sleep


[86b] Leverziekten. H.L.A. Janssen, J.P.H. Drenth, B. van Hoek. Boek; 1e druk; 08-10-2009; 9789031374366. 2.5 0


Huang Di Nei Jing: Su Wen, Uit Engelse bronnen vertaald door Guus Martens ISBN 109086660142


Sun Z , Jia J, Gong X et al. (2012). Inhibition of glutamate and acetylcholine release in behavioral improvement induced by electroacupuncture in parkinsonian rats. Neurosc. letters. 520(1); pp 32-37.


Biophysical journal vol100, 1910-1918.


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