Review Article

Influence of Chemical Elements on Mammalian Spermatozoa

(sperm / spermatozoa / semen / chemical element / trace elements / heavy metals / motility / capacitation / acrosome reaction / oxidative stress / male infertility)

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Abstract. Exposure to heavy metals is the most important risk factor in the assessment of spermatogenesis. About 30–40 % cases of infertility are caused by the male factor, and most of them are due to the small quantity of spermatozoa or to inferior spermatozoa quality. The negative impact on sperm motility, morphology and concentration of such chemical elements as Al, Cr, Cd, Pb or Fe was observed, while positive influence was noticed for Zn, Mg, and Ca. The influence of Mn, Cu, Ni or Se on spermatozoa is ambiguous. Chemical elements known as necessary for capacitation and acrosome reaction are Zn, Mg and Ca, while Cd and Pb disturb initiation and progress of the acrosome reaction. The positive effect of chemical elements Al, Cd, Cr, Cu, Ni, Pb, Se, and Zn, lies in their protection against oxidative stress. On the other hand, Al, Cu and Ni induce structural changes in the testes and epididymis or influence interactions with other chemical elements.

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Abbreviations: CASA – computer-assisted semen analyser, CAT – catalase, FSH – follicle-stimulating hormone, HMW-Zn% – Zn-high-molecular-weight proteins, IVF – *in vitro* fertilization, SOD – superoxide dismutase, WHO – World Health Organization.

Introduction

Natural environmental factors and differentiated anthropogenic pollutants, as well as many other sources strongly influence the reproductive material located in the semen, both in animals and humans. Chemical elements constitute an important group of ecophysiological influence among these sources. These elements disturb semen material and they are divided into several groups (Simkiss, 1975; Underwood, 1977; Fergusson, 1990):

1/ Most important and essential elements for semen (highest physiological role: Na, K, Ca, Mg and Fe).
2/ Trace elements, i.e. microelements, with their concentration in semen required for its proper (normal) functioning in relatively narrow limits, on a limited scale (i.e. we can note the relatively narrow ranges of tolerance of these elements, with concentration in "excess" and "deficiency", which is toxic for semen). Most of them are heavy metals, e.g. Zn, Cu, Mn and Co. Selenium as a microelement can also be included into this group, since it is known to interact and may be toxic itself (Hoffman et al., 1985).
3/ Toxic heavy metals; even a trace amount in semen is very harmful and dangerous. The main such metals are Pb and Cd, though Hg is also included here. These metals are defined as elements with characteristic lustrous appearance, they are good conductors of electricity, and generally enter chemical reactions as positive ions or cations.

However, with regard to the semen quality and condition, sometimes the distinction between metals and non-metals is not sharp. E.g., Sb, As and Te have physical properties of metals and chemical properties of non-metals. Generally, the distinction between metals and non-metals is even less clear and depends very much on personal prejudice. Similarly, the distinction between metals that are heavy and those that are not is often unregistered. Many authors define metals as heavy if they have a relative density greater than 4.5 g × cm^{-1}, but there are many papers in which this term is used to de-

scribe elements that are neither heavy (e.g. Al), nor are metals in the strict sense (e.g. Se). The term "trace metals" is also difficult to define. In studies on vertebrates, metals are described as being present in trace amounts if their concentrations in the tissue are lower than that of Fe (Underwood, 1977).

The requirement for particular elements is demonstrated by comparing growth, survival and reproductive success of a control group with a group fed on a diet that is identical except for the absence of the element under test. A number of criteria have been established which must be satisfied before an element can be considered as essential for semen, i.e.: 1/ removal of the element from the diet should result in disturbed growth and reproduction; 2/ these deficiency effects should be accompanied by pathological changes and status of immunoactivity; 3/ alleviation of deficiency effects should be dose-dependent; 4/ the element should form an essential component of an antioxidant enzyme, hormone or another biologically active substance (Hopkin, 1989). Many elements considered to be essential for vertebrates have satisfied only the first criterion. However, it may be impossible to disprove an essential requirement for some elements as the theoretical lowest possible limit for essentiality is one atom per one specific gene in the chromosomes of a cell. The distinction between essential and non-essential elements is therefore equivocal (Hopkin, 1989).

Exposure to heavy metals is a risk factor in the assessment of spermatogenesis. Biogeochemical management of chemical elements enables their transfer to the ecological system, accumulation in the soil, ground waters, and then accumulation in plant and animal organisms, moving between all links of the trophic chain that ends in human. Generally, ordinary human diet ensures sufficient quantity and proper proportions of trace elements necessary for correct development and health. Trace elements essential for animals and human As, B, Br, Cl, Co, Cu, F, Fe, I, Li, Mn, Ni, Se, Sn, V and Zn are also important for the semen quality. Chemical elements essential for semen quality in very small concentrations (ng/kg) and/or by their undefined function are Ba, Cd, Pb, Rb, Sr and Ti. Depending on their concentration in the organism, trace elements can cause disturbances of biochemical homeostasis. However, the reproductive epithelium is one of the most sensitive tissues reacting upon any changes of biogeochemical and environmental parameters (Kabata-Pendias and Pendias, 1999; Kurpisz, 2002; Semczuk and Kurpisz, 2006; Kabata-Pendias and Mukherjee, 2007).

Besides chemical elements with their various ecophysiological impact, many other chemical pollutants, contaminators, and chemical drugs influence human and animal semen. They all reduce sperm quality in mammals, including humans. Among these aromatic hydrocarbons, polychlorinated biphenyls, chlorofluorocarbons, bromofluorocarbons, pesticides, a large group of endocrine disrupters, drugs and other pollutants play an important role. However, we must also emphasize that the ecotoxicological impact on the semen quality often differs because of the different biomodels and the differences in sperm maturation; namely the difference between ejaculated (human) and epididymal sperm (mouse), various doses of test substances, their applications, and differences between *in vivo* and *in vitro* experiments with various mammalian sperm.

The influence on the motility, morphology and concentration of spermatozoa

The motility, morphology and sperm concentration are the basic parameters in standard estimation of semen. This examination is at present the most important clinical test for estimation of fertility. About 30–40% of the causes of infertility are associated with the male factor (Vayena et al., 2002); the most of them are due to the small quantity of spermatozoa or to their inferior quality. Oligozoospermia might be the cause of infertility in accordance with the World Health Organization (WHO) criteria for men with less than 20 million sperm/ml of semen. The decrease in the sperm count can be caused by various reasons. However, it generally occurs at the final stage of spermatogenesis. Appropriate sperm motility and morphology is necessary for fertilization of the oocyte besides the spermatozoa quantity. Spermatozoa with abnormal morphology are not capable to fertilize the oocyte. Usually, the disturbances of morphology testify to abnormal genetic material of the spermatozoon (Pisarski and Szamatowicz, 1997; Kurpisz, 2002).

The negative impact on the motility, morphology and concentration of spermatozoa of such chemical elements as Al, Cr, Cd, Ni, Pb or Fe was observed. Aluminium is suspected as one of the environmental sources of pollution liable to decrease the sperm quality. In studies of the sperm quality of Finnish men it was shown that a high concentration of Al was correlated with decreased sperm motility and disturbed sperm morphology. Moreover, patients with the highest Al concentration in their spermatozoa (from 8.7 to 21.5 mg/kg) showed symptoms of asthenozoospermia (Hovatta et al. 1998). Rabbits receiving the dose of $AlCl_3$ at 34 mg/kg body weight displayed decreased sperm concentration, sperm motility and ejaculate volume (Yousef et al., 2005).

Chromium in the human organism is distributed rather regularly in each tissue and is necessary for its normal development. In the experiments with monkeys (*Macaca radiata* Geoffrey), chromium given in drinking water for six months caused reduction of sperm counts and sperm forward motility (Subramanian et al., 2006). Additionally, laboratory mice injected with CrO_3 at doses of 1 mg/kg of body weight displayed increased rates of sperm abnormalities (Acharya et al., 2006). Simultaneously, decreased sperm counts and percentages of motile sperm (Li et al., 2001) as well as increased percentages of morphologically abnormal spermatozoa were found in men occupationally exposed to Cr(VI) (Kumar et al., 2005).

Cadmium is easily absorbable, relatively long accumulated in the tissues conducting essential functions in the organism, and represents a special risk for human health. Cd toxicity impairs the reproductive functions of living organisms (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). The elevated levels of Cd were found in azoospermic subjects, and the negative impact of Cd was shown in all examined biophysical semen characteristics except for the sperm volume (Akinloye et al., 2006). At the same time, positive correlations were observed between Cd and sperm motility, and Cd and both linear velocity and curvilinear velocity (analysed by computer-assisted videomicrography) (Noack-Fuller et al., 1993). In the experiments with 2 mM CdCl solution, the sperm motility first decreased, and in the third hour of incubation the velocity and amplitude of the movement suddenly increased and then again considerably decreased (Battersby et al., 1982). The studies by Massányi et al. (2004) suggested that Cd has a direct negative effect on spermatozoa quality, especially on the sperm morphology. The same authors reported the occurrence of separated flagellum of spermatozoa caused by Cd (Massányi et al., 2005). In oligoasthenozoospermic subjects, negative correlations between Cd concentration in the sperm and sperm motility and sperm concentration (density) were found (Pant et al., 2003). However, there is also a report in which no significant differences in Cd concentration between the sperm of teratozoospermic, asthenozoospermic and oligozoospermic subjects and normozoospermic subjects were found (Kasperczyk et al., 2002).

The negative impact (separated flagellum of spermatozoa) was also suggested for Ni (Massányi et al., 2004; Zemanová et al., 2007). Nickel is found in all tissues and its effects are not manifested immediately because up to 90 % of the received Ni is accumulated in bones (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Higher seminal fluid Pb concentrations were also observed in infertile (3.6 ± 3.2 μg/l) than in fertile men (1.7 ± 1.0 μg/l) (Saarenen et al., 1987), and its negative impact on the motility and viability of spermatozoa and sperm count was observed (De Rosa et al., 2003; Eibensteiner et al., 2005). A significant increase in the percentage of abnormal spermatozoa was found in rats given intraperitoneal injections of 25 mg/kg of Pb acetate or 25 mg/kg of Pb acetate with 4 mg/kg of Zn acetate (Piao et al., 2007). However, some reports have not found any influence of Pb on the sperm (Butrimovitz et al., 1983; Noack-Fuller et al., 1993; Jackenhovel et al., 1999).

Iron and iron compounds are not essentially toxic for animals and human organisms. Nevertheless, disturbances in the regulative absorption mechanism can appear due to pathological conditions or prolonged intake of high Fe doses. In these cases Fe is bound in the form of ferric phosphate (haemosiderin) or into proteins, and is distributed in the liver (Kabata-Pendias and Pendias, 1999; Semczuk and Kurpisz, 2006; Kabata-Pendias and Mukherjee, 2007). The increased Fe concentration can also bear negatively on the morphology of spermatozoa (Massányi et al., 2004); significant differences in the Fe concentration between sperm of severely teratospermic subjects were reported in contrast to no differences in normozoospermic subjects (Kwenang et al., 1987).

The positive effects on the motility, morphology and concentration of spermatozoa were reported for Zn, Mg, Ca and Ni. The deficit of Zn is also associated with disturbances of reproductive functions. Zinc is present both in spermatozoa and in seminal plasma, where its concentration is considerably higher than in the other body fluids. Zinc also contributes to the form of sperm motility. It was noticed that seminal fluid with higher percentage of motile spermatozoa contains plasma with higher Zn concentration (Kaludin et al., 1983; Wong et al., 2001). Zinc in immature spermatozoa is mainly located in outer dense fibres of the flagellum, where it is bound to the sulphhydryl groups of cysteine. The majority of its content is reduced during epididymal sperm maturation, which leads to increased stabilization of outer dense fibre proteins by oxidation of sulphhydryl groups to disulphide bridges. This stabilization of outer dense fibre proteins seems to be an essential step for the generation of sperm motility, especially progressive motility (Henkel et al., 1999). The studies by Carpino et al. (1998) suggest that the increased content in the free Zn unbound to high molecular proteins (HMW-Zn%) in seminal plasma is associated with decreased percentage of motile spermatozoa in asthenozoospermic patients. Moreover, Zn in seminal plasma occurs in lower concentration in asthenozoospermic and oligoasthenozoospermic patients (Zhao and Xiong, 2005), while men with low Zn levels in the blood serum are more exposed to the risk of asthenozoospermia (Yuyan et al., 2008). The high Zn concentration also has inhibitory influence on the progressive motility of spermatozoa, but not on the percentage of motile spermatozoa (Sørensen et al., 1999). The direct influence of Zn on sperm morphology was also suggested (Massányi et al., 2004), because increased percentages of broken flagellum with the decreased Zn content were noticed (Massányi et al., 2005). According to Kumar et al. (2006), in experiments with zinc-supplement-fed bulls, higher semen volume, sperm concentration, percentage of live sperm and motility were found. Additionally, higher percentages of motile spermatozoa were reported in semen with higher concentration of Mg in seminal plasma (Wong et al., 2001).

Calcium is required in many physiological processes as a regulator in all living cells, including spermatozoa. Although the influence of Ca on motility has not been completely elucidated, it was noticed that the addition of Ca^{2+} with calsemin to isolated ram caudal spermatozoa caused stimulation of flagellar beat activity (Bradley and Forrester, 1982). On the other hand, negative correlation between Ca concentration in seminal plasma and motility of spermatozoa was found in bull spermatozoa (Machal et al., 2002).

The influence of Mn, Cu or Se on spermatozoa is ambiguous. Copper is an essential element for the normal

function of the organism. Copper deficiency causes decreased fertility (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Copper can also have negative impact on the morphology of spermatozoa (Gamik et al., 1990; Massányi et al., 2004, 2005; Ramamoorthi et al., 2008) and sperm motility (Gamik et al., 1990). The incubation of spermatozoa in the presence of Cu had a negative effect on some of motility parameters (distance and velocity) examined by computer-assisted semen analyser (CASA) in studies by Roychoudhury et al. (2008) and caused decreased sperm motility or complete sperm immotility (Maynard et al., 1975; Battersby et al., 1982; Holland and White, 1982). On the other hand, positive influence of Cu on sperm concentration and counts was also noticed. For example, men with sperm concentrations above 40 million/ml showed higher Cu semen concentrations than men with azoospermia, displaying sperm concentrations below 5 million/ml and between 10 and 20 million/ml (Jackenhovel et al., 1999). Positive correlation was noticed between Cu concentration in blood, sperm count in the ejaculate, and count of spermatozoa with progressive motility, and between Cu concentration in seminal plasma and the volume of ejaculate, motility of spermatozoa and number of spermatozoa with progressive motility (Machal et al., 2002). However, no significant differences in the effect of Cu concentration were found in teratospermic and normozoospermic men (Kwenang et al., 1987). The high Mn level was associated with decreased sperm motility and concentration (Wirth et al., 2007). However, other reports demonstrated that seminal fluid with higher percentage of motile spermatozoa contained plasma with higher concentration of Zn and Mn (Kaludin et al., 1983).

Selenium is generally associated with amino acids, cysteine (selenocysteine) and methionine (selenomethionine) in living organisms. Simultaneously, as it was indicated in experiments by Saarenen et al. (1987), higher Se concentrations were observed in the blood plasma of infertile men. Selenium appears in higher concentration in semen than in seminal plasma and affects sperm concentration and percentage of normally formed sperm (Noack-Fuller et al., 1993; Hawkes and Turek, 2001; Akinloye et al., 2005). This element increased motility of spermatozoa in the presence of Cr (Ramamoorthi et al., 2008). Saarenen et al. (1989) also noticed correlations between low sperm Se content and abnormal morphology and motility of bull spermatozoa. In the mouse both excess and deficiency of Se in the diet caused reduction in sperm concentration and motility (Shalini and Bansal, 2008). On the other hand, Wirth et al. (2007) have not found any Se impact on any of the examined quality parameters.

The effects on the capacitation process and induction of acrosome reaction

Capacitation is a temporary and reversible stage of maturation of the male gamete, which prepares the spermatozoon to fertilization except for changes in the cell membrane. The results of capacitation are modifications of the track form and type of sperm motility (Semczuk and Kurpisz, 2006). Chemical elements known to be required in capacitation and acrosome reaction are Zn, Mg and Ca. Zinc plays an important role in capacitation e.g. as a regulatory factor (Andrews et al., 1994; Bilaspuri and Babbar, 2007). Andrews et al. (1994) noticed reduction of the Zn level in the acrosomal region during capacitation (by 44 % or 40 %), but this did not correlate with the occurrence of spontaneous acrosome reactions. Magnesium takes part in various metabolic processes such as formation of DNA and RNA structures, lipid management, and activation of many enzymes (KabataPendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Magnesium and calcium maintain the osmolar balance and take part in nutrient transfer. The presence of Mg^{2+} and Ca^{2+} ions is necessary for the last stage of capacitation, and the following acrosome reaction and hyperactive motility of spermatozoa (Semczuk and Kurpisz, 2006). Experiments have shown that addition of ionophore A23187, which actively transports Ca^{2+} ions from extracellular to intracellular space, induces the acrosome reaction (Aitken et al., 1993). However, it was proved that the initiation of acrosome reaction with the ionophore is possible in the presence of Ca^{2+} ions in the extracellular space (De Jonge, 1994). There are also experiments in which exposure of capacitated spermatozoa to periovulation follicular fluid or progesterone caused calcium influx from extracellular space and initiation of acrosome reaction (De Jonge, 1999).

The chemical elements particularly modulating the initiation and duration of acrosome reaction are Cd and Pb. The negative impact of Cd on male fertility consists in its chemical affinity to Ca channels in the cell membranes and decreased ability of spermatozoa to undergo acrosome reaction (Benoff et al., 2000). Arabi and Mohammadpour (2006) also discovered that Cd may alter the integrity of acrosomal membranes of spermatozoa and cause abnormal acrosome reaction. Lead is suspected for inexplicable infertility of men caused by induction of spontaneous premature acrosome reaction, and its content in seminal plasma may have a negative impact on *in vitro* fertilization (IVF) rates (Benoff et al., 2003).

Induction of oxidative stress, histological changes and element-element interactions

The semen quality and the ability of spermatozoa to undergo capacitation and acrosome reaction are not the only factors influenced by chemical elements and contributing to infertility. The effect of these elements also consists in the protection against oxidative stress or its modification, induction of changes in the activity of acrosomal membrane and epididymis, or interactions with other chemical elements. Experiments in the mouse (*Mus musculus*) revealed that Al caused reduction of

testicular and epididymal weight and decrease of spermatid count. The spermatid count was reduced at an intraperitoneal dose of 100 mg/kg/day of body weight and at 200 mg/kg/day of body weight led to histological changes and significant decrease of testicular weight (Llobet et al., 1995). In rabbits that received the $AlCl_3$ dose of 34 mg/kg of body weight, a decrease in libido (by increasing the reaction time), live body weight, feed intake and weight of testes and epididymis were observed (Yousef et al., 2005). In the experiments with incubation of rabbit spermatozoa in $AlCl_3$ solution at the concentrations of 10, 15 and 20 mM, significant induction of oxidative stress and inhibition in the activities of superoxide dismutase (SOD) and catalase (CAT) were noticed (Yousef et al., 2007).

Chromium can bind to nucleic acids and plays an essential role in the metabolism of glucose, some proteins and lipids. Chromium is an important component of enzymes and stimulates their mutual activity. Deficiency of Cr occurs rarely (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). In the studies by Aruldhas et al. (2005) and Subramanian et al. (2006) done on monkeys (*Macaca radiata*) that received drinking water containing Cr(IV), a negative Cr impact on spermatogenesis mediated by induction of oxidative stress was observed. Chromium treatment also disrupted spermatogenesis, leading to accumulation of prematurely released spermatocytes and spermatids in the lumen of seminiferous tubules. Granulation of chromatin and vacuolation between the acrosomal cap and manchette microtubules of elongated spermatids and in the Golgi area of round spermatids were observed (Pereira et al., 2005). The specific activities of antioxidant enzymes (superoxide dismutase, catalase, glutathione peroxidase, glutathione reductase and glucose-6-phosphate dehydrogenase) and non-enzymatic antioxidants (glutathione, vitamins A, C and E) decreased, whilst the concentration of H_2O_2 and hydroxyl radicals in the testes increased. Induction of oxidative stress in the experiments with mice receiving Cr(VI) were also noticed (Pereira et al., 2005). In men occupationally exposed to Cr(VI), decreases of Zn concentration in the sperm and increases of follicle-stimulating hormone (FSH) in the blood serum were observed (Li et al., 2001).

Zinc, as a component and activator of various enzymes, also plays a significant role in the metabolism of proteins and carbohydrates, and probably lipids. Zinc with magnesium also stabilize cell membranes (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Additionally, Zn with Fe take part in the processes of oxidation and reduction, and Zn together with Cu prevent deleterious effects of reactive oxygen species on spermatozoa as cofactors of Cu-Zn superoxide dismutase (Gavella and Lipovac, 1998; Kabata-Pendias and Pendias, 1999; Semczuk and Kurpisz, 2006; Kabata-Pendias and Mukherjee, 2007). Zinc in P2 protamines may play a role in stabilization of sperm chromatin and in inhibition of transcription (Bianchi et al., 1992). Zinc deficiency may lead to degeneration changes in the cells involved in spermatozoa processing after meiotic division (Cigankova et al., 1998). It was shown that the semen pH increase was caused by the decrease of Zn concentration in seminal fluid (Krupej et al., 1994).

Cadmium is suspected for diminishment of male fertility through the binding to the structure of sperm chromatin (Casswell et al., 1987). The incubation of spermatozoa with Cd caused a fall of sperm nuclear Zn concentration (Battersby et al., 1982). Accumulation of Cd by spermatozoa caused a significant decrease in the protein level and increase in catalase activity.

Manganese takes part in various physiological processes, especially as activator of enzymes regulating the metabolism of glucose, other carbohydrates, lipids including cholesterol, and proteins. Manganese is usually not the component of these enzymes. The function of this metal is not specific and it can be replaced by other metals, especially by Mg. Manganese occurring in the cells is found mainly in the mitochondria and binds to DNA and RNA. The decrease of fertility connected with Mn deficiency is a secondary effect of the disturbance of cholesterol synthesis and related chemical compounds which are necessary for synthesis of sex hormones and other steroids. Manganese excess can impair the metabolism of other chemical elements and limit their bioavailability. A large quantity of Mn mostly induces deficiency of Cu, P and Fe (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Ramamoorthi et al. (2008) noticed that Mn decreased the percentage of motile spermatozoa in the presence of Cu. Copper occurs in many enzymes of oxidative-reductive processes, takes part in the absorption of Fe and is a cofactor of superoxide dismutase, which prevents deleterious effects of reactive oxygen species on spermatozoa (Semczuk and Kurpisz, 2006). Incubation of spermatozoa in Cu-containing solution caused a fall in Mg concentration in the nucleus and acrosome regions (Battersby et al., 1982) and a decrease of Na content in the head and mid-piece, as well as a decrease of K and Zn content in the head while augmenting the Cu level (Maynard et al., 1975). Copper with zinc decreased the quantity of glucose utilized by spermatozoa and the quantity of glucose oxidized, and caused the accumulation of lactate (Holland and White, 1980). Vrzgulová et al. (1995) also observed a negative impact of Cu on the microscopic structure of the testes. The accumulation of Cu by spermatozoa caused a significant decrease in the protein level and increase in the catalase activity (Formicki et al., 2007).

Nickel is an activator of some enzymes (dehydrogenases and carboxylases). At high concentrations it is harmful, both for animals and humans, causing e.g. changes in chromosomes and neoplastic cytogenesis. Nickel excess influences the metabolism of other metals, primarily decreasing the level of Mg and of Mn and Zn in some parenchymal organs (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Research in rats revealed that $NiCl_2$ induced shrinkage

of seminiferous tubules and decreased the number of basal spermatogonia in the testes (Kakela et al., 1999). Nickel deficiency can also have a negative impact on spermatogenesis and semen quality. In the experiment with rats supplemented by dietary Ni at 1 mg/kg, Yokoi et al. (2003) found a significant decrease in the density of epididymides, epididymal transit time of spermatozoa, testes sperm production rate, and weight of seminal vesicles and prostate. Studies in mice revealed that testicular toxicity of Ni may be related to enhanced production of reactive oxygen species, which leads even to DNA damage (Doreswamy et al., 2004).

Interactions between Pb and other chemical elements might essentially disturb the metabolism of chemical elements necessary for health and semen quality. Increased Pb concentrations enhance excretion of Fe and Cu. The increase of Cu level in the diet decreases the sorption of Pb. The association between Pb and Se consists in a secondary effect of synthesis of poorly solvable Pb selenides, which accumulate in the kidneys. Calcium and phosphorus also have an antagonistic effect on Pb (Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Benoff et al. (2000) also reported a negative Pb impact on male fertility in view of its chemical affinity to K channels, while Robins et al. (1983) observed a possible damaging effect of Pb on the chromatin of spermatozoa and spermatogenesis. The high exposure to Pb most likely impaired spermatozoa through increased lipid peroxidation (Kasperczyk et al., 2008).

Selenium plays a significant role in human organism as a component of glutathione peroxidase, thereby protecting the organism against the damaging effect of hydrogen peroxides and hydroxyl radicals of lipids. Because generation of metal (e.g. Hg, Pb, Ag, Ta) selenides in the human organism, which for their poor dissolubility are liable to exclusion from biochemical processes, occurs easily, Se can immobilize the toxically active excess of metals that accumulate mostly in parenchymal organs; however, this may appear disadvantageous for the general metabolism (Pasternak and Floriańczyk, 1995; Kabata-Pendias and Pendias, 1999; Kabata-Pendias and Mukherjee, 2007). Shalini and Bansal (2008) also observed that in laboratory tests of mice treated by dietary Se deficiency, the spermatozoa had incompletely condensed chromatin and increased occurrence of DNA strand breaks, while both dietary Se excess and deficiency caused reduction of fertilization.

Iron participates in the processes of oxygenation and reduction, entering into composition of many enzymes and metalloprotein compounds. Generally, the elementary function of Fe in the cells is protection against toxic products of oxygenation reactions. Both, absorption and metabolic function of Fe are linked with influence of other chemical elements. Particularly antagonistic activity is exerted by Cd, Mn, Pb and Zn. Interactions with Cu are complex and frequently synergetic during their cooperation in the oxidation and reduction processes. Although Fe and its compounds are not toxic for animals and human organisms, its overload can result in increased sperm DNA damage (Perera et al., 2002).

Previous studies of the influence of each chemical element on spermatozoa frequently demonstrated conflicting results. The knowledge in this area is still incomplete and demands complementation, although many results of current research define the impact of particular elements on spermatozoa or explore the sperm quality of men occupationally exposed to heavy metals. Definition of appropriate levels of these elements seems of particular importance, as well as the contribution of particular elements to particular fertility parameters. For example, conclusive answers concerning the activities of Cu, Mn and Se on the motility, morphology and concentration of spermatozoa should be determined. The chemical elements participating in capacitation and acrosome reaction are Zn, Mg and Ca, whilst the initiation and acrosome reaction processes are disturbed by Cd and Pb. Protective effects against induction of oxidative stress were noticed for Al, Cd, Cr, Cu, Ni, Pb, Se, and Zn, whilst histological changes were caused by Al, Cu and Ni (Pasternak and Floriańczyk, 1995; Kabata-Pendias and Pendias, 1999; Semczuk and Kurpisz, 2006; Kabata-Pendias and Mukherjee, 2007).

Based on previous and recent investigations in the field of chemical elements' impact on the quality and condition of semen, we can conclude that although trace elements play an essential role in spermatogenesis and fertility, the determination of total seminal concentration of these elements may not be a useful tool for determining the sperm fertilization potential. Some heavy metals might bear negatively on the morphology of spermatozoa and the determination of their level in semen during infertility investigation is thus recommended. The activities of antioxidant enzymes, particularly superoxide dismutase, are related to the parameters of human semen and their monitoring may be a useful tool for determining the sperm fertilization potential and could improve the diagnosis of male infertility. The conflicting results of some previous studies on the influence of trace elements on spermatozoa and the relation of antioxidant enzymes activity with the parameters of human semen should be elucidated by further investigations.

The fact that chemical radicals, particularly trace elements and toxic heavy metals, play an important role in spermatogenesis and fertility and might bear negatively on the morphology of spermatozoa is well known. However, the relationships between trace metals and the function of sperm production and sperm motility are still unclear. Further studies should therefore be done, focusing also on other elements, enzymatic and non-enzymatic mechanisms, and other contributing proteins.

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