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*Comparison of the levels of copper, zinc and iron in cervical mucus
and in blood serum of women of childbearing age*

The role of trace elements in various body fluids is the issue of general interest. Trace elements serve their key function as a cofactor for many enzymes. They are indispensable in many intracellular biological processes (10), they are among the essential components necessary for the proper functioning and integrity of the immune system (14), and they also make a part of the mechanisms for getting rid of free radicals (12). The majority of research studies are focused on blood serum as a carrier delivering trace elements to various organs and tissues, and on the medium of the enzymatic reactions requiring the presence of trace elements (1, 4, 11, 12). Levels of zinc, copper and iron in blood serum reflect the amount of these elements in food supply (1). More and more often the role of these elements in food intake is discussed and advocated (8, 7). The deficiency of these elements may induce functional disturbances of many systems and/or organs (8, 13).

The available literature on the presence of trace elements in cervical mucus is rather scant (2). Cervical mucus is a mixture of the secretion of the mucosa of cervical canal, uterine cavity, uterine tubes and the fluid produced by peritoneum (9). The daily production of the mucus changes in quantity (6) and it significantly increases in periovular period, when it amounts to about 600 mg per day (5). *In vitro* studies have proved the great impact of trace elements on sperm function in the cervical mucus medium (3), and the so-called cervical factor is one of the most important causes of infertility. Almost all copper pool of the cervical mucus can be found in the mucinous fraction while zinc can be found, first of all, in the watery fraction (2). Zinc plays multifunctional role in the mucus, on the one hand it is indispensable in order to preserve the proper defense reaction in this medium (14), and on the other hand it can participate in sperm capacitation which takes place, among others, in the uterine cervix.

MATERIAL

Levels of iron, copper and zinc were determined in cervical mucus and blood serum of ten patients of childbearing age. None of them were pregnant at the time of the research studies. They took no drugs whatever during three months preceding the studies, they had no intrauterine devices and none of them received hormonal contraceptives. All of the patients were duly informed about the investigation aim, and they gave their informed consent to the investigation.

METHOD

The mucus samples were collected during a routine gynaecological examinations when patients were in their periovular period. Plastic disposable colposcopes and tuberculinic syringes were used for sampling. The collected material was stored at a temperature of -20°C . Simultaneously blood samples were also collected in order to determine the levels of trace elements in serum. After the formation of coagulum the blood samples were centrifuged at 2000 rpm for 20 minutes. Then blood serum was transferred to some other new plastic test-tubes and frozen at a temperature of -20°C .

The sample material was weighted and placed in test-tubes of well known quantity. The samples were divided into three groups and mineralized in supra pure concentrated nitric acid. The microwave generator with three-step cycle and power rise (60%, 80%, and 100%) was used. This step led our substance into solution. After mineralisation and neutralization the material was analyzed chromatographically with the use of 500DX Dionex with derivatisation on-line and 530 nm wave length. Quantity analysis was prepared with calibration curve. All dilutions were prepared with redistilled water and purified in Eas-Pure device. Purity was examined conduct metrically.

The obtained results were statistically analysed. The Statistica 5.0 computer program was used for the research studies. The association between parameters was examined with the use of Spearman Rank Order Correlation R Test and in order to find out the differences between the examined characteristics the Mann-Whitney U test was used.

OBJECTIVE

The objective of the research study was to preliminarily compare concentrations of trace elements in cervical mucus and in blood serum of women of childbearing age.

RESULTS

The average value of iron (Fe) in mucus was 2.99 mg/g (range 0.89 mg/g to 8.25 mg/g), and in blood serum it was 0.51 mg/g (range 0.37 mg/g to 3.18 mg/g). The value of copper (Cu) was respectively 0.17 mg/g in mucus (range 0.07 mg/g to 0.93 mg/g) and 0.44 mg/g in serum (range 0.24 mg/g to 0.99 mg/g), while zinc (Zn) was 0.12 mg/g in mucus (range 0.02 mg/g to 0.48 mg/g), and 0.54 mg/g in serum (range 0.11 mg/g to 0.62 mg/g).

No correlation was found between the investigated trace elements in cervical mucus and in blood serum there was only a positive correlation found between iron and copper ($R = 0.75$; $p = 0.01$).

No statistically significant correlation was found between the trace elements in cervical mucus and in blood serum.

DISCUSSION

The relevant literature concerning the contents of trace elements in cervical mucus is very scant. There are some reports on concentration of zinc in the bovine cervical mucus during normal oestrus and pharmacologically induced oestrus (15). These reported values are several times higher than the values obtained in our present studies (15). While examining the contents of trace elements in women cervical mucus, D a u n t e r (2) divided mucus into fractions: a "watery" fraction which was formed after adding physiological saline and a "mucinous" fraction received after centrifugation of the sample material and removal of the watery fraction. Danter's study concerned the follicular and the luteal phases and they found no differences in these phases as far as the iron and zinc levels were concerned, and the contents of copper in the watery fraction increases slightly in the luteal phase. Our present

studies are focused on periovular period and cervical mucus as a whole, without dividing it into fractions. Daunter's studies showed that almost the whole copper quantity can be found in the "mucinous" fraction, which can be associated with its bonding with mucus albumins, while iron and zinc can be found exclusively in the watery fraction (2). The above interdependence may be linked with the viscosity of cervical mucus and its crystallisation. Tsiligianni (16) suggests the relation between the amount of trace elements in the mammal cervical mucus and viscosity, and shows negative correlation with crystallisation. He associates the water content in cervical mucus with the trace element concentrations.

It is difficult to compare the results of our studies with those of other researchers because of the different form of sampling and the different method of determination of trace elements concentrations in body fluids (ASA vs. chromatography) and also because of a different moment of sampling in the menstrual cycle.

No comparative studies on trace elements in cervical mucus and blood serum were found in the available professional literature. Our study did not reveal any correlation between the levels of zinc, copper and iron in cervical mucus and in blood serum.

CONCLUSIONS

As no correlation was found between the blood serum and cervical mucus, as far as trace elements are concerned, one might suggest that the uptake of trace elements by the mucus has an active form.

REFERENCES

1. Abdulla M., Suck C.: Blood levels of copper, iron, zinc, and lead in adults in India and Pakistan and the effect of oral zinc supplementation for six weeks. *Biol. Trace Elem. Res.*, 61(3), 323, 1998.
2. Daunter B.: Trace metals (Cu, Mn, Zn, Fe), sulphhydryl and disulphide groups of cervical mucus. *Contraception*, 15, 543, 1977.
3. Eggert-Kruse W. et al.: Microbial colonization and sperm-mucus interaction: results in 1,000 infertile couples. *Human Reprod.*, 7, 612, 1992 (a).
4. Forrer R. et al.: Simultaneous measurement of the trace elements Al, As, B, Be, Cd, Co, Cu, Fe, Li, Mn, Mo, Ni, Rb, Se, Sr, and Zn in human serum and their reference ranges by ICP-MS. *Biol. Trace Elem. Res.*, 80, 77, 2001.
5. Insler V., Bettendorf G.: *The uterine cervix in reproduction*. G. Thieme Verlag, Stuttgart 1977.
6. Insler V., Lunenfeld B.: *Diagnose und Therapie endokriner Fertilitätsstörungen der Frau*. Bd.11. Grosse-Verlag, Berlin 1977.
7. Iyengar G.V., Nair P.P.: Global outlook on nutrition and the environment: meeting the challenges of the next millennium. *Sci. Total Environ.* 249, 1-3, 331, 2000.
8. Klevay L. M.: Lack of a recommended dietary allowance for copper may be hazardous to your health. *J. Am. Coll. Nutr.* 17, 4, 322, 1998.
9. Moghissi K.: The cervix in infertility *Clin. Obstet. Gynecol.*, 22, 1, 27, 1979.
10. Pena M. et al.: A delicate balance: Homeostatic control of copper uptake and distribution. *J. Nutr.*, 129, 1251, 1999.
11. Perveen S. et al.: Effect of gestational age on cord blood plasma copper, zinc, magnesium and albumin. *Early Hum. Dev.* 69, 1-2, 15, 2002.

12. Rukgauer M. et al.: The relation between selenium, zinc and copper concentration and the trace element dependent antioxidative status. *J. Trace Elem. Med. Biol.*, 15, 2-3, 73, 2001.
13. Semrad C. E.: Zinc and intestinal function. *Curr. Gastroenterol. Rep.*, 1, 5, 398, 1999.
14. Sturniolo G. C. et al. Altered plasma and mucosal concentrations of trace elements and antioxidants in active ulcerative colitis. *Scand. J. Gastroenterol.*, 33, 6, 644, 1998.
15. Tsiligianni T., Karagiannidis A., Brikas P., Saratis P.: Physical properties of bovine cervical mucus during normal and induced (progesterone and/or PGF₂ α estrus). *Theriogenology*, 55, 629, 2001.
16. Tsiligianni T., Karagiannidis A., Roubies N. et al.: Concentration of calcium, zinc, magnesium, potassium and sodium in the bovine cervical mucus during normal oestrus and oestrus induced by progesterone and/or PGF₂ α . *Repr. Fertil. Dev.*, 14, 427, 2002.

SUMMARY

The objective of this research study was a preliminary comparison of some chosen trace elements concentrations in cervical mucus and blood serum of women of childbearing age. The levels of iron (Fe), copper (Cu) and zinc (Zn) were determined in cervical mucus and serum of 10 patients. The obtained results do not point to any association between the levels of the examined trace elements in cervical mucus or any correlation between their contents in mucus and blood serum. As no correlation was found between the blood serum and cervical mucus, as far as trace elements are concerned, one might suggest that the uptake of trace elements by the mucus has an active form.

Porównanie stężeń miedzi, cynku i żelaza w śluzie szyjkowym i w surowicy krwi kobiet w wieku rozrodczym

Celem pracy było wstępne porównanie stężeń wybranych pierwiastków w śluzie szyjkowym i w surowicy krwi kobiet w wieku rozrodczym. Oznaczono poziomy Fe, Cu, Zn w śluzie szyjkowym i surowicy krwi u 10 pacjentek. Uzyskane wyniki nie wskazują na to, aby istniał związek pomiędzy poziomami badanych pierwiastków w śluzie szyjkowym oraz pomiędzy ich zawartością w śluzie i surowicy krwi. W związku z niezalezieniem zależności między surowicą krwi a śluzem szyjkowym można domniemywać, iż sposób pozyskiwania pierwiastków w śluzie odbywa się w sposób aktywny.