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**INFLUENCE OF BENTONITE ON ABSORPTION OF ZINC  
SUPPLEMENTS IN RATS**

**WPŁYW BENTONITU NA WCHŁANIANIE U SZCZURÓW  
ZWIĘKSZONYCH ILOŚCI CYNKU W DIECIE**

**Key words:** bentonite, supplemental zinc, absorption, rat.

**Słowa kluczowe:** bentonit, cynk, suplementacja, wchłanianie, szczur.

*Badania wykonano na dwóch grupach samców szczurzych szczepu Wistar. W grupie I (kontrolnej) zwierzęta spożywały standardową paszę LSM, a w grupie II tę samą paszę wzbogaconą w 2-procentowy dodatek bentonitu. W obu grupach zwierzęta otrzymywały przez 28 dni dożołądkowo w wodzie pitnej chlorek cynku (około 23 mg/l) znakowany cynkiem 65, tak by ilość cynku podawana zwierzętom była dwukrotnie większa od zalecanej. Zawartość cynku 65 oznaczano radiometrycznie w korpusie z usuniętym przewodem pokarmowym w okresie do 28 dni po zaprzestaniu aplikacji. Wyniki wskazują, że dodatek bentonitu zwiększał zawartość cynku w organizmie zwierząt. Biorąc pod uwagę wartości parametru AUC, wzrost ten wyniósł ponad 50%. Dane mogą mieć praktyczne znaczenie tam, gdzie stosowane są pasze wzbogacone w bentonit, a zawartość cynku w diecie zwierząt jest podwyższona.*

**1. INTRODUCTION**

Bentonite is a clay comprising minerals resulted from in situ devitrication of volcanic ash. Its name was derived from the Fort Benton cretaceous rocks in Wyoming (US) where it was first found in about 1890. The composition of bentonite may vary according to its geographical location but as a mixture of minerals it has not any chemical formula. It comprises mainly hydrated aluminium silicates smectite minerals such as montmorillonite (named after the town Montmorillon in central France) and nontronite. Bentonite has strong colloidal

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properties and it swells to about 12 to 15 times its volume when coming into contact with water. Moreover, bentonite has great adsorptive capacity; one gram of bentonite has a surface area of about 800 sq. meters. These properties of bentonite make it a valuable material for a wide range of uses and applications [Abehsera 1979, McGraw-Hill encyclopedia 1988, Trickova et al. 2004]. The beneficial action of bentonite consists in the adsorption of toxins and inclusion of bacteria and certain viruses such as intestinal influenza present in the gastrointestinal tract and trace elements including iron, calcium, selenium, and manganese [Abehsera 1978, Grosicki et al. 2004 a, b, Grosicki, Rachubik 2005, 2009, Schwartz, Werner 1990].

Little is known about the influence of bentonite on zinc absorption and metabolism. It was reported that a prolonged oral application of high doses of bentonite reduced zinc incorporation into the liver and kidneys of goats [Schwarz and Werner, 1990]. In contrast, supplemental bentonite given to rats enhanced moderately the absorption and organ retention of zinc given in traces amounts [Grosicki, Rachubik 2009] Increases in zinc absorption following bentonite additives may be undesirable especially in the areas with high zinc concentrations. Thus, the present paper attempts to examine the influence of bentonite on the absorption and distribution of zinc given at a dose higher than zinc requirements in rats.

## 2. MATERIALS AND METHODS

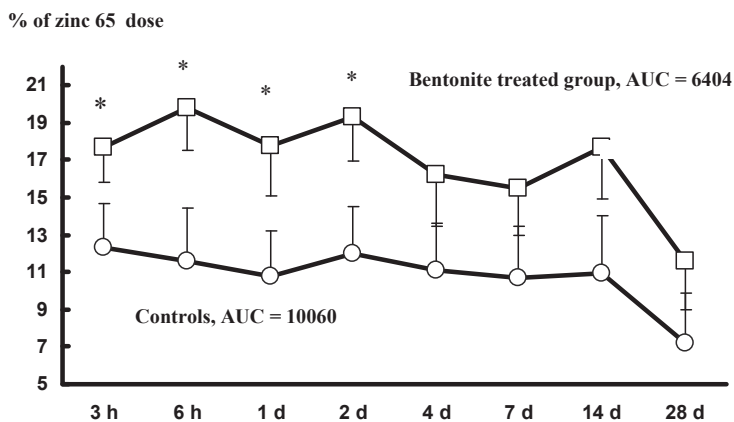
Ninety male Wistar rats weighing  $207\text{g} \pm 11\text{ g}$  were used. The animals were randomly assigned into two dietary groups each of 45 rats after an acclimatisation period of one week. Rats in the two groups (controls and bentonite treated) were offered tap water fortified with 23 mg Zn/L and a standard rodent chow LSM *ad libitum* (Fodder Manufacture Motycz, Poland). The bentonite treated group received the same LSM diet but fortified with 2% of bentonite. The total zinc content of the LSM diet was 23.3 mg/kg according to the manufacturer. The bentonite used originated from the Polish geological sources. The animals were on these diets for the whole experimental period. Body weight gains and feed and water consumption were recorded weekly during the feeding period.

Animals received chloride in a 0.5 mL water solution comprising about 20 kBq of zinc 65 per rat daily for 28 d except weekends by an intragastric tube. Rats were killed by immersion in gaseous carbon dioxide 3 h, 6 h, 1 d, 2 d, 4 d, 7 d, 14 d, and 28 d after dosing. Radiozinc in the carcass (whole body without the stomach and intestines) was measured using a whole-body counter ZM 701 (Polon, Poland). Reference standards for quantification of carcass radiozinc were prepared by intraperitoneal injection of the appropriate solution to rats, which were killed 45 min thereafter.

The area under the curves (AUC) of radiozinc content versus time points was calculated by the trapezoidal rule. Data were analysed statistically using Student's *t*-test at  $P < 0.05$ .

### 3. RESULTS

The consumption of feed, body weight gain, relative weight of liver and kidneys were similar in all tested rats and no statistically significant differences between the two groups were noted (data not shown). Moreover, these parameters were not different from those noted in rats fed the LSM diet without bentonite.



\* – means significant differences at  $P < 0.05$ .

**Fig.** Distribution of zinc 65 in the carcass of rats

**Rys.** Zawartość cynku 65 w korpusach szczurów

As shown in Fig., the comparison of zinc 65 carcass retention in the controls and bentonite treated groups shows differences between the two groups examined. The fortification of the diet with 2% of bentonite increased carcass retention of zinc in comparison to that found in the controls. The increases persisted throughout the whole experimental period and were statistically significant within the first 2 days postdosing. The content of zinc 65 in the carcass expressed as AUC values indicated that the absorption of zinc in rats fed the bentonite fortified diet increased by about 57%.

### 4. DISCUSSION

No statistically significant differences in organ to body ratios in the rats fed the bentonite enriched diet indicated that a 2% dietary supplement of bentonite seemed to be not toxic to rats, at least over the course of 28 experimental days. However, there are reports indicating that higher concentrations of bentonite (from 10% to 50%) may influence unfavourably body

weight gains in animals [Adamis et al. 2005]. The effect of bentonite on the bioavailability of zinc seems to be controversial [Grosicki, Rachubik 2005, Schwartz, Werner 1990] and may depend on the amount of bentonite applied to diet. Higher dietary concentrations of bentonite [from 10% to 50%] decreased hepatic and renal zinc content whereas a concentration as low as 2% increased zinc concentrations in selected organs of rats [Grosicki, Rachubik 2009, Schwartz, Werner 1990]. The results of the present report provided evidence that bentonite used as a 2% dietary supplement increased zinc bioavailability even when a zinc administration doubled the requirements for this element. It is rather difficult to explain why bentonite considered an agent with high adsorbent capability [Adamis et al. 2005, Ma, Uren 1998, Trckova et al. 2004] increases zinc uptake from the gastrointestinal tract. The effect of bentonite on trace element bioavailability seems to be complex. Findings reported by several authors showed that bentonite may affect absorption of several trace elements making them more or less available for the utilization by animals [Abdel-Wahhab et al. 2002, Grosicki et al. 2004, Grosicki, Rachubik 2009, Schwartz, Werner 1998].

The findings showing an increased zinc uptake from the alimentary canal of animals fed supplements of bentonite indicate that this agent may play a significant role in zinc utilization by animals. It may pose particular problems in cases where zinc concentrations in foodstuffs is high as a results of environmental contamination.

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