Vol. 11 (2002): 137–141.

The effect of clinoptilolite on ^{137}Cs binding in broiler chickens

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The objective of this study was to evaluate the ^{137}Cs binding capacity of clinoptilolite. In the first *in vitro* experiment we investigated sorption of ^{137}Cs to natural and modified forms of clinoptilolite in highly acid solution, prepared to be similar to that of the gut of pigs (pH = 2–3) at 37°C. In the second *in vivo* experiment ^{137}Cs binding to a modified form of clinoptilolite was studied in orally contaminated broiler chickens. ^{137}Cs sorption in the high acidity solution depended on clinoptilolite concentration and varied between 30–85% of the initial activity. In the chickens, three hours after ^{137}Cs administration, there was 67% and 63% lower accumulation of ^{137}Cs in meat and edible organs (respectively) and seven hours after ^{137}Cs administration, there was 69% and 49% lower accumulation of ^{137}Cs in meat and edible organs (respectively) compared to the controls with no clinoptilolite added in food. Natural and modified forms of clinoptilolite have been shown to high sorption efficiency towards ^{137}Cs ions and could be recommended as possible radiocaesium binders in domestic animals.

Key words: clinoptilolite, ^{137}Cs, sorption, broilers

Introduction

Contamination of the environment with radioactive substances has become one of the most important problems in modern civilization. Therefore, there is a need for constant and systematic control of radionuclide concentrations in the biosphere. Radioactive contamination of the environment as a consequence of the Chernobyl nuclear accident has led to contamination of vegetation and feedstuffs for animal nutrition with biologically significant radionuclides, ^{90}Sr, ^{131}I and ^{137}Cs. Prevention of contamination via the oral ingestion of these radionuclides is a priority in radioprotection. In animal production, there is a need to ensure that contamination of animal products is below intervention levels.

Manuscript received May 2001

Howard et al. (2001) reviewed the current availability of countermeasures for preventing or reducing contamination of animal products by radioisotopes of iodine, caesium and strontium.

Reducing contamination via the gut of domestic animals and the consequent health risk for humans, requires the development of methods which use binding substances to bind radiocaesium in the digestive tract of animals and eliminate it through faeces. After the first nuclear accidents, initial investigations were conducted on ^{137}Cs sorption. Potassium was added into the feed to stimulate radiocaesium elimination from the organism (Williams and Patrick 1957). Beresford et al. (1989) used bentonite as a radiocaesium binder in sheep and achieved a reduction of between 20–80%. Currently, the best caesium binding effects have been achieved by Prussian Blue compounds, the most commonly used compound being AFCF - ammonium ferric hexacyanoferrate (Arnaud et al. 1988, Hove and Ekern 1988, Unsworth et al. 1989, Hove and Hansen 1993, Paasikallio et al. 2000). Reduction of 90–98% was achieved in cows with 3 g of AFCF daily, calves and pigs 2 g of AFCF daily (Giese 1988) and in broilers (Vitorovic 1993) with 0.2 g of AFCF daily per chick. Philippo et al. (1988), Pethes et al. (1988) and Jandl and Novosad (1995) demonstrated that zeolite (a clinoptilolite clay mineral) can be used as a possible binder for ^{137}Cs in the gut of domestic animals.

Zeolites are crystalline, hydrated aluminosilicates that have an indefinite three-dimensional structure. Their characteristic is the ability to exchange water and some of its constitutional cations without significant changes in their structure. Natural zeolites are used as catalysts in many reactions, as molecular sieves and for cation exchange (Ming and Mumpton 1989). Considering this, zeolites have wide use as animal feed additives with the ability for mycotoxine adsorption (Rodrigues 1991, Harvey et al. 1993, Amon and Dobesic 1994, Krilov et al.1994). Clinoptilolite could be also used in agriculture to reduce ^{137}Cs deposition in soil because of its ability to bind ^{137}Cs (Paul and Jones 1995, Campbell and Davies 1997, Jones et al. 1999).

The objective of the present study was to evaluate the ^{137}Cs binding capacity of a natural white form of clinoptilolite and different modified clinoptilolite forms in *in vitro* and *in vivo* conditions.

Material and methods

In the *in vitro* experiment, natural clinoptilolite from Zlatokop mine was used. The investigated zeolite had above 80% clinoptilolite mainly in Ca(II) form. Silicate analysis was used for Si and Al determination, atomic absorbtion for Na, K, Ca, Mg determination and the Kjeldhal method for NH_4^+ ion. The cation exchange (Na, K, Ca and Mg) capacity was above 130 cmol kg^{-1}. In the first stage of this experiment, 100 mg and 300 mg of clinoptilolite were treated with 100 ml electrolytic solution of HCl, NaCl and KCl, to which 1440 Bq ^{137}Cs was added. The acidity of the solutions was initially adjusted to be pH 2–3, with NaOH, which resembles the acidity condition within the gut of pigs. Sorption was performed in a water bath at 37ºC with slow shaking. Contact times were 2, 4, 6 and 8 hours and three replicates were taken for each period of time. After sorption, the samples were centrifuged and remaining activity measured in the separated liquid fraction. In the second stage of this experiment, we compared ^{137}Cs binding capacity of natural and mono-ionic, sodium (Na) and ammonium (NH_4) forms of clinoptilolite. The hydrated samples of natural clinoptilolite were treated with 3M NH_4Cl and 3M NaCl solutions to prepare the mono-ionic forms of clinoptilolite. 500 mg of the sample was equilibrated with 50 ml electrolytic solution to which 1440 Bq ^{137}Cs was added under the same solution composition and acidity as in the first stage of the experiment. The contact time was 6 hours at 37ºC after which the remaining activity of ^{137}Cs in solution was measured, as before.

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For the *in vivo* experiment, broiler chickens (six weeks of age) were fed with their normal diet and each chick was orally given a 3 ml CsCl solution with a total activity of 3120 Bq. In addition, the experimental group was given a 6 ml aqueous solution of clinoptilolite modified with NaCl called Minazel (1.6 g Minazel per chick which is approximately 1–1.5% in the diet). Principles of laboratory animal keeping were in accordance with national laws for animal protection and welfare. Birds of each group were sacrificed 3 and 7 hours (five chicks per group) after the contamination was administered. Samples of total musculature and edible organs (heart, gizzard and liver) were taken from each chick. A HP Ge detector with high-energy resolution (1.85 KeV – 1.33 MeV) was used for gamma spectrometery measurement of activity of the samples. The measurement time was 40,000 s. The effect of clinoptilolite treatments on ^{137}Cs binding was evaluated by analysis of variance and differences between treatments were tested using t-test.

Results and Discussion

Administration of the lower clinoptilolite concentration (100 mg) reduced by half the initial ^{137}Cs activities (52%) in solution during 6 hours of incubation. Prolongation of the contact time gave no further decreasing in solution activity (48.6% for 8 hours). Using a higher clinoptilolite amount (300 mg), the equilibrium of Cs ions was also established after 6 hours, but 75% (74.7% in 6 hours and 74.2% in 8 hours) of initial ^{137}Cs activities was sorbed. The higher clinoptilolite administration therefore have a higher sorption efficiency (between 67.6–74.7%) than that of the lower amount (30.4–48.6 %). The activities of solution depending of different contact times and clinoptilolite concentrations are given in Table 1.

Natural and prepared forms of clinoptilolite (Na- and NH_4- clinoptilolites, 500 mg) showed similar and significant effects on ^{137}Cs sorbtion, sorbing 84–85% of initial activity (Table 2). The higher ^{137}Cs sorption efficiency of clinoptilolite in this experiment compared to the results in the first stage was the result of using higher clinoptilolite concentrations. Philippo et al. (1988) established that clinoptilolite and hexacyanocobalt ferrate are the most effective adsorbers of radiocaesium *in vitro*. When used *in vivo* they significantly reduced the absorption of radiocaesium by sheep fed contaminated herbage.

In our *in vivo* experiment, ^{137}Cs binding efficiency was evaluated for the modified clinoptilolite form - Minazel, in broiler chickens. Three hours after administering ^{137}Cs contamination and the mineral, significanly ($P < 0.01$) lower ^{137}Cs activity concentration was obtained in the

Table 1. Clinoptilolite efficency in sorption of Cs^{137} from a solution, with an activity of 1440 Bq.

Time of contact (hours)	^{137}Cs activity in solution (Bq) 100 mg clinoptilolite Mean	SD	300 mg clinoptilolite Mean	SD	Sorbed activity (% of starting activity) 100 mg clinoptilolite	300 mg clinoptilolite
2	1001.0	15.0	466.0**	10.0	30.4	67.6
4	894.0	4.0	383.0**	8.0	37.8	78.4
6	748.0	15.0	364.0**	11.0	48.0	74.7
8	739.0	12.0	371.0**	10.0	48.6	74.2

SD = standard deviation

** = statistically significant differences ($P < 0.01$) between treatment of clinoptilolite concentration.

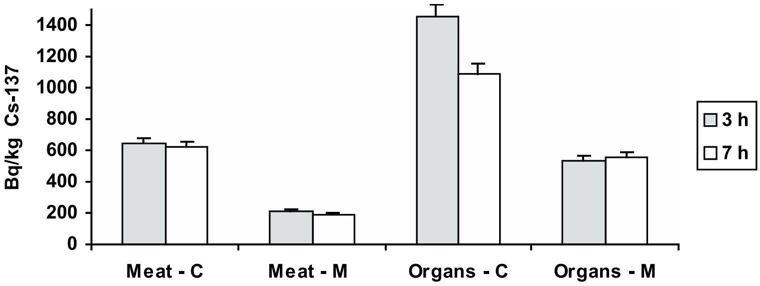

Fig. 1. Efficiency of Minazel in reducing ^{137}Cs accumulation in the meat and edible organs of broiler chickens, orally contaminated with 3150 Bq ^{137}Cs per chick, abbreviation C is for “control” and M for “Minazel” (means and standard errors).

meat of broilers treated with Minazel than in the control group. Compared to the control group, the reduction of ^{137}Cs accumulation in the treated group was 67%. Seven hours after contamination and mineral administration, the decrease due to ^{137}Cs binder was 69%. Pethes et al. (1988) found that accumulation of radiocaesium was significantly decreased by adding 6% zeolite to broiler feed. The accumulation of ^{137}Cs in zeolite treated groups ranged 62–70% of the non-treated groups. Jandl and Novosad (1995) investigated *in vivo* reduction of radiocaesium in sheep using modified clinoptilolite (ZEOFe) and non-modified clinoptilolite (ZEO). More than ten times lower sorption efficinecy has been observed with ZEO comparison to ZEOFe. In our investigation, the modified clinoptilolite Minazel, showed significant protection effect in edible organs (heart, gizzard and liver) of broilers contaminated with ^{137}Cs. Three hours after contaminaton and mineral administration there was about 63% lower accumulation of ^{137}Cs in edible organs of the group treated with Minazel compared to controls; after seven hours it was 49%.

Considering that these are preliminary results in our investigation of the ability of clinoptilolite to bind ^{137}Cs in the digestive organs of domestic animals, it is still too early to give suggestions for its practical application and to compare it with other ^{137}Cs binders (bentonite, vermiculite, AFCF). It is also too early to evaluate animal products obtained using zeolite as ^{137}Cs binder in human nutrition especially because in this investigation the degree of oral ^{137}Cs contamination of chickens (3150 Bq per day) was far above potential radioactive contamination of chick feeds in accidental situations (like the one produced by Chernobyl). It was used to obtain a clear data on the efficiency of clinoptilolite in ^{137}Cs binding in the digestive tract of chickens. In our further investigations more attention will be given to radioactive contamination of animal feeds in practical conditions with long-term observations in the ability of clinoptilolite to bind ^{137}Cs in orally contaminated animals.

Table 2. Efficiency of different forms of clinoptilolite in ^{137}Cs sorption from a solution, with an activity of 1440 Bq.

Forms of clinoptilolite	^{137}Cs activity (Bq) Mean	SD	Sorbed activity (% of starting activity)
Natural	230.0	11.0	84.0
Sodium (Na-)	215.0	11.0	85.1
Ammonium (NH_4-)	215.0	7.0	85.3

SD = standard deviation

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Conclusion

Natural clinoptilolite and modified forms of clinoptilolite have a high sorption efficiency towards ^{137}Cs ions. In Serbia, there are deposits of this particular mineral of high quality. Because of that, and its low price, it could be recommended as a possible radiocaesium binder in domestic animals. Additional *in vivo* studies are needed to evaluate optimal clinoptilolite concentrations for different species, breed and categories of domestic animals.

Acknowledgements. We thank Mrs Jelena Perovic, from Patentcommerce, Belgrade, official distributor of clinoptilolite and Minazel, for providing help in this investigation.

References

Amon, M. & Dobesic, M. 1994. Influence of local clinoptilolite in the broiler chickens production. *Proceeding of the 8th International Congress of Animal Hygiene.* St. Paul, Minnesota, USA. p. 210–217.

Arnaud, M.J., Clement, C., Getaz, F., Tanhauser, F., Schonegge, R., Blum, J. & Giese, W. 1988. Synthesis, effectiveness and metabolic fate in cows of the caesium complexing compound ammonium ferric hexacyanoferrate labelled with ^{14}C. *Journal of Dairy Research* 55: 1–13.

Beresford, N.A., Lamb, C.S., Mayers, R.W., Howard, B.J. & Colgrove, P.M. 1989. The effect of treating pastures with bentonite on the transfer of ^{137}Cs from grazed herbage to the sheep. *Journal of Environmental Radioacitivty* 9: 251–264.

Campbell, L.S. & Davies, D.R. 1997. Experimental investigation of plant uptake of caesium from soils amended with clinoptilolite and calcium carbonate. *Plant and Soil* 189: 1, 65–47.

Giese, W.W. 1988. Ammonium-ferric-cyano-ferrate (II) (AFCF) as an affective antidote against radicaesium burdens in domestic animals and animal derived foods. *British Veterinary Journal* 144: 363–369.

Harvey, R.B., Kubena, L.F., Elisade, M.H. & Phillips, T.D. 1993. Efficiacy of zeolitic ore compounds on the toxicity of aflatoxin to growing broiler chickens. *Avian Dissease* 37: 67–73.

Hove, K. & Ekern, A. 1988. Combating radiocaesium contamination in farm animals. *Health problems in connection with radiation from radioactive matter in fertilizers, soils and rocks.* J. Log. Oslo, Norwegian University Press. p. 139–153.

Hove, K. & Hansen, H. 1993. Reduction of radiocaesium transfer to animal products using sustained release boli with ammoniumiron (III) – hexacyanoferrate (II). *Acta Veterinaria Scandinavica* 34: 287–297.

Howard, B., Beresford, N. & Voigt, G. 2001. Countermeasures for animal products: a review of effectiveness and potential usefulness after an accident. *Journal of Environmental Radioactivity* 56: 115–137.

Jandl, J. & Novosad, L. 1995. In vivo reduction of radiocaesium by modified clinoptilolite in sheep. *Veterinaria Medicina* 40: 237–241.

Jones, D.R., Paul, L. & Mitchell, N.G. 1999. Effects of ameliorative measures on the radiocaesium transfer to upland vegetation in the UK. *Journal of Environmental Radioactivity* 44, 1: 55–69.

Krilov, M.P., Fantin, V.M., Kumarin, S.V., Krilova, N.I. & Sadykov, S.M. 1994. Zeolites in feed mixtures for cows. *Zootekhnia* 10: 12–14.

Ming, D.W. & Mumpton, F.A. 1989. Zeolites in Soils Environment. In: *Minerals in Soil Environment.* Soil Science Society of America, Madison, Wisconsin, USA. p. 873–909.

Paasikallio, A., Sormunen-Cristian, R., Jaakkola, S. & Kaikkonen, M. 2000. The effect of ammonium ferric hexacyanoferrate on reducing radiocaesium transfer from grass silage to sheep. *Agricultural and Food Science in Finland* 9: 135–147.

Paul, L. & Jones, D.R. 1995. Effects of ameliorative treatmens applied to radicaesium contaminated upland vegetation in the United Kingdom. In: *Environmental impact of radioactive releases*. Proceedings of Symposium, Vienna, Austria. p. 513–522.

Pethes, G., Rudas, P., Bartha, T. & Frenyo, V.L. 1988. Accumulation and elimination of radiocaesium consumed in fodder from different tissues of broiler chickens and rabbits. *Acta Veterinaria* 38: 299–306.

Phillippo, M., Gvozdanovic, S., Gvozdanovic, D., Chesters, J.K., Paterson, E. & Mills, C.F. 1988. Reduction of radiocaesium absorption by sheep consuming feed contaminated with fallout from Chernobyl. *Veterinary Record* 122: 560–563.

Rodrigues, J.R. 1991. Use of sorghum with zeolite or sugar in diets for broiler chickens. *Revista Cubana de Cienca Avicola* 18, 2: 144–149.

Unsworth, E.F., Pearce, J., McMurray, C.M., Moss, B.V., Gordon, F.J. & Rice, D. 1989. Investigations of the use of clay minerals and Prussian Blue in reducing the transfer of dietary radiocaesium to milk. *The Science of the Total Environment* 85: 339–347.

Vitorovic, G. 1993. The application of ammonium-ferric (III)-hexa-cyanoferrate (II) in the protection of chicken meat from contamination with radiocaesium. *Acta Veterinaria* 43: 351–358.

Williams, R. & Patrick, H. 1957. Metabolism of Cs-137 in rabbits. *Archive of Biochemistry and Biophysics* 70: 464–468.