# USE OF Na-CITRATE, Ca-GLUCONATE, AND HEXACYANOFERRATES IN REDUCING RADIOCAESIUM CONTAMINATION IN RATS

## ANDRZEJ GROSICKI AND BOGDAN KOWALSKI

Laboratory of Radiological Protection and Isotope Studies, National Veterinary Research Institute, 24-100 Pulawy, Poland

Ammonium ferric ferrocyanoferrate (AFCF), Na-citrate and Ca-gluconate or their combination were assessed for their efficacy in reducing the whole-body, carcass and organ contaminations with <sup>137</sup>CsCl given intragastrically for 5 consecutive days. The most advantageous effect was obtained when 10 doses of AFCF were used. Similar results were produced following the combined treatment with AFCF plus Na-citrate and Ca-gluconate. Fivefold injections of Na-citrate and Ca-gluconate without AFCF treatment caused no effect on mobilizing Cs-137 from the body and organs of rats.

Radiocaesium is the principal radionucleide of health significance for man and animals and as such has attracted much attention. The health effect of radiocaesium is attributed to a long physical and biological half-life and its accumulation in plants and food-producing animals (5, 11, 13).

Removal of radiocaesium from the environment is a complex problem and involves, among others things, both the soil-to-plant (9) and plant-to-animal transfers (2). In the latter, the radiocaesium transfer to animals is dramatically decreased by inhibition of the intestinal absorption and enhancement of bodily elimination as a result of hexacyanoferrate and clay mineral application into the alimentary tract (1, 3, 6, 8, 9, 16). However, a serious limitation of these agents is that their effectiveness is profoundly reduced if treatment is delayed (10).

In the case of an accidental radiocaesium release into the environment a large proportion of this radionucleide is taken up by animals which consequently leads to contamination of people consuming food of animal origin. It is, therefore, of importance to reduce internally deposited radiocaesium in animals. Success in mobilizing caesium-137 depends on the use of the agents forming a soluble caesium-agent complex which is effectively excreted from the body.

Hackett (4), Ryabova (14), Smith et al. (15), and Volf (17) using citrate and gluconate substantially increased radiocaesium, radiostrontium, and radioplutonium elimination from the animal body. However, no attention has been paid to the radiocaesium mobilization from organs after the therapy.

The object of the present experiment is to evaluate the decontamination effectiveness of Na-citrate and Ca-gluconate in rats contaminated with caesium-137. Moreover, ammonium ferric ferrocyanoferrate of known role in radiocaesium

elimination and its combination with citrate and gluconate were used to evaluate the relative effect of these treatments.

#### **Materials and Methods**

The animals used were male Wistar rats weighing about 250 g. Rats were allowed free access to a commercial pelleted chow (LSM, Bacutil, Poland) and tap water. Caesium chloride (137CsCl, Polon, Poland) was administered by intragastric tube to rats for 5 consecutive days at a daily dose of 20 kBq in a volume of 0.6 ml of distilled water.

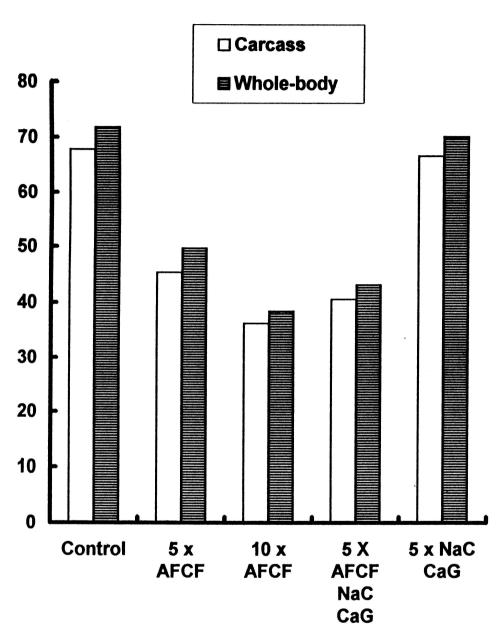
Animals were killed 6 days after termination of the experimental protocol. The whole-body and carcass retentions of Cs-137 were measured in a well type scintillation counter ZM-701 (Polon, Poland). The blood, liver, kidneys, brain and muscle samples were collected and weighed and the Cs-137 contents were determined in a ZR-11 scintillation counter (Polon, Poland).

A reference standard for quantification of whole-body and carcass radioactivities was prepared by ip injection of the appropriate solution of caesium-137 into 5 rats killed 30 min thereafter. Statistical evaluation was based on 10 rats per group and mean values were compared by Student's t-test.

### Results

The doses of AFCF and Na-citrate and Ca-gluconate produced no effect on the body weight gain and organ/body ratios of the liver, kidneys, heart and testicles in any group investigated. Macroscopic examinations showed no evidence of organ lesions. The whole-body and carcass retentions of caesium-137 from intragastric doses are shown in Fig.1. The controls retained in the body and carcass 71.77 and 67.75% of the caesium-137 doses, respectively, after fivefold administration.

Fivefold or tenfold treatment with AFCF or fivefold treatment with AFCF plus 5 injections of Na-citrate and Ca-gluconate (Groups 2 through 4) decreased significantly the whole-body retention of radiocaesium up to 49.66%, 38.41%, and 43.17%, respectively. Five injections of Na-citrate and Ca-gluconate (Group 5) revealed no effect (70.07%) on the whole-body retention compared to that in the controls. There were no significant differences among Groups 2 - 4.



NaC - sodium citrate CaG - calcium guconate

Fig. 1. Whole-body and carcass retentions of caesium-137 (per cent of dose).

The carcass retention pattern of radiocaesium in Groups 2 through 5 closely resembled that of the whole-body retention. There were only slight reductions in the carcass Cs-137 contents with respect to the corresponding values of the whole-body retention.

The contents of Cs-137 in the blood, liver, kidneys, muscles, and brain are listed in Table 1. For the controls, a maximum concentration of Cs-137 after fivefold dosing was found in the liver followed by kidneys, muscles, brain, and blood. A similar pattern of radiocaesium distribution among the organs and tissues was observed in Groups 2 through 5.

In the kidneys, muscles, and brain the greatest reduction of caesium-137 level was found after the tenfold AFCF administration; the combined treatment was the most effective in reducing caesium-137 level in blood and muscles. The fivefold treatment with AFCF was less effective in reducing radiocaesium in the brain and muscles in comparison to that resulting from the tenfold AFCF and combined therapy. Treatments with AFCF and the combined one produced statistically significant decreases in caesium-137 level in all organs, examined except the brain and muscles, in rats given 5 doses of AFCF. No statistically significant differences were found among Groups 2 - 4.

Fivefold Na-citrate and Ca-gluconate injections were ineffective in mobilizing caesium-137 from the organs of rats. Surprisingly, the burden with Cs-137 in muscles, blood, and brain was even increased when compared to that in the controls. In contrast, renal and hepatic Cs-137 levels decreased slightly in comparison to the corresponding values in the controls. However, these alterations were statistically insignificant.

### Discussion

The lack of deleterious effects of the applied Na-citrate and Ca-gluconate on rat growth rate, organ/body ratios, and health is in agreement with the results of others (10). The general pattern of radiocaesium distribution among the rat organs and tissues resembles that reported in an earlier study (8).

The present results clearly indicate that the efficacy of tested chemicals in promoting caesium-137 removal from rat body and major sites of its sequestration was related to the chemical agents and dosages involved. With the exception of Nacitrate and Ca-gluconate therapy, decreases in caesium-137 contents followed by the AFCF and combined treatments were significant statistically. A high effectiveness of AFCF confirmed earlier studies with animals (1, 6, 7, 8, 16). It seems, however, that the decontamination effectiveness would be even more remarkable if the delay between contamination and the beginning of therapy was as short as possible in order to prevent the transfer of caesium-137 from the gastrointestinal tract (10).

In general, our findings failed to confirm earlier observations of Ryabova (10) on the substantial increase in caesium-137 elimination from animal bodies after citrate and gluconate injections. The results obtained indicate that Na-citrate and Cagluconate produce an increase in the decontamination effectiveness of AFCF therapy but when given alone these chelates reveal no effect at all on caesium-137 mobilization from the rat body.

Table 1

Effect of AFCF, Na-citrate, and Ca-gluconate on Cs-137 content in rat organs and tissues.

5×NaC 5×CaG 5×CaG 4* 0.025±0.004* 0.0 54.35% 15* 1.572±0.302* 2.0 52.04% 52.04% 54.49% 0.388±0.091* 0.0 54.49% 54.49% 0.0 54.49% 0.0 54.49% 0.0 54.49% 0.0 54.49% 0.0 54.49% 0.0 54.49% 0.0 54.49% 0.0 54.49% 0.0		Control	5×AFCF	10×AFCF	5×AFCF	5×NaC
5×Cs-137       5×Cs-137       5×Cs-137       5×Cs-137         0.046±0.006       0.029±0.006*       0.027±0.004*       0.025±0.004*         100%       63.04%       58.69%       54.35%         3.000±0.460       1.981±0.471*       1.55±0.345*       1.572±0.302*         100%       66.03%       51.73%       52.04%         ys       0.712±0.069       0.452±0.086*       0.317±0.045*       0.388±0.091*         es       0.262±0.044       0.178±0.034       0.135±0.021*       0.160±0.025*         100%       67.94%       51.52%       61.06%         0.078±0.011       0.060±0.007       0.046±0.012*       0.057±0.010*         100%       76.92%       58.97%       73.07%					5×NaC	5×CaG
5×Cs-137       5×Cs-137       5×Cs-137       5×Cs-137       5×Cs-137         0.046±0.006       0.029±0.006*       0.027±0.004*       0.025±0.004*       0.025±0.004*         100%       63.04%       58.69%       54.35%       54.35%         3.000±0.460       1.981±0.471*       1.552±0.345*       1.572±0.302*         100%       66.03%       51.73%       52.04%         52.04%       66.03%       63.17±0.045*       0.388±0.091*         100%       63.48%       44.52%       54.49%         es       0.262±0.044       0.178±0.034       0.135±0.021*       0.160±0.025*         100%       67.94%       51.52%       61.06%         0.078±0.011       0.060±0.007       0.046±0.012*       0.057±0.010*         100%       76.92%       58.97%       73.07%					5×CaG	
0.046±0.006       0.029±0.006*       0.027±0.004*       0.025±0.004*         100%       63.04%       58.69%       54.35%         3.000±0.460       1.981±0.471*       1.552±0.345*       1.572±0.302*         100%       66.03%       51.73%       52.04%         ys       0.712±0.069       0.452±0.086*       0.317±0.045*       0.388±0.091*         ys       0.262±0.044       0.178±0.034       0.135±0.021*       0.160±0.025*         es       0.262±0.044       67.94%       51.52%       61.06%         0.078±0.011       0.060±0.007       0.046±0.012*       0.057±0.010*         100%       76.92%       58.97%       73.07%		5×Cs-137	5×Cs-137	5×Cs-137	5×Cs-137	5×Cs-137
100% 63.04% 58.69% 54.35% 54.35% 3.000±0.460 1.981±0.471* 1.552±0.345* 1.572±0.302* 1.00% 66.03% 51.73% 52.04% 52.04% 51.73% 52.04% 51.00% 63.48% 44.52% 54.49% 54.49% 51.52±0.044 0.178±0.034 0.135±0.021* 0.160±0.025* 100% 67.94% 51.52% 61.06% 61.06% 100% 76.92% 58.97% 73.07%	Blood	0.046±0.006	0.029±0.006*	0.027±0.004*	$0.025\pm0.004*$	0.052±0.023
3.000±0.460 1.981±0.471* 1.552±0.345* 1.572±0.302* 100% 66.03% 51.73% 52.04% 52.04% 52.04%  95 0.712±0.069 0.452±0.086* 0.317±0.045* 0.388±0.091* 100% 65.348% 44.52% 54.49% 54.49% 51.452% 61.06±0.025* 100% 67.94% 0.135±0.021* 0.160±0.025* 100% 0.078±0.011 0.060±0.007 0.046±0.012* 0.057±0.010* 100% 76.92% 58.97% 73.07%		100%	63.04%	28.69%	54.35%	113.04%
ys 0.712±0.069 66.03% 51.73% 52.04% 100% 66.03% 63.17±0.045* 0.388±0.091* 0.312±0.069 0.452±0.086* 0.317±0.045* 0.388±0.091* 0.262±0.044 0.178±0.034 0.135±0.021* 0.160±0.025* 0.078±0.011 0.060±0.007 0.046±0.012* 0.057±0.010* 0.057±0.010* 0.060±0.007 0.046±0.012* 0.057±0.010* 0.057±0.010* 0.060±0.007 0.046±0.012* 0.057±0.010* 0.057±0.010* 0.057±0.010* 0.057±0.010* 0.057±0.010* 0.060±0.007 0.057±0.010* 0.057* 0.057* 0.050* 0.050* 0.050* 0.050* 0.050* 0.050*	Liver	$3.000\pm0.460$	$1.981\pm0.471*$	$1.552\pm0.345*$	$1.572\pm0.302*$	$2.672\pm0.284$
ys 0.712±0.069 0.452±0.086* 0.317±0.045* 0.388±0.091* 100% 63.48% 44.52% 54.49% es 0.262±0.044 0.178±0.034 0.135±0.021* 0.160±0.025* 100% 67.94% 51.52% 61.06% 0.078±0.011 0.060±0.007 0.046±0.012* 0.057±0.010* 100% 76.92% 58.97% 73.07%		100%	%60.99	51.73%	52.04%	89.07%
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es 0.262±0.044 0.178±0.034 0.135±0.021* 0.160±0.025* 100% 67.94% 51.52% 61.06% 61.06% 0.078±0.011 0.060±0.007 0.046±0.012* 0.057±0.010* 100% 76.92% 58.97% 73.07%		100%	63.48%	44.52%	54.49%	87.64%
$100\%$ $67.94\%$ $51.52\%$ $61.06\%$ $0.078\pm0.011$ $0.060\pm0.007$ $0.046\pm0.012*$ $0.057\pm0.010*$ $100\%$ $76.92\%$ $58.97\%$ $73.07\%$	Muscles	$0.262\pm0.044$	$0.178\pm0.034$	$0.135\pm0.021*$	$0.160\pm0.025*$	$0.269\pm0.023$
0.078±0.011 0.060±0.007 0.046±0.012* 0.057±0.010* (100%) 76.92% 58.97% 73.07%		100%	67.94%	51.52%	61.06%	102.67%
76.92% 58.97% 73.07%	Brain	$0.078\pm0.011$	0.060±0.007	$0.046\pm0.012*$	$0.057\pm0.010*$	0.082±0.009
		100%	76.92%	58.97%	73.07%	105.12%

\* - significant differences, P<0.05; NaC - sodium citrate; CaG - calcium gluconate.

Smith (15) in a report on the use of citrate to increase the removal of radiostrontium from animals suggested that metabolic blocking of the citrate-strontium complex within the renal tubules should enhance strontium excretion from the body. Moreover, it was found that in the case of radiostrontium contamination citrate is effective only when injected shortly after contamination with radiocaesium.

The injection of citrate and gluconate one hour after contamination was in accordance with the procedure described by Ryabova (10). Thus, in the view of Smith's findings the ineffectiveness of citrate and gluconate therapy in our experiment may eventually result from a long time interval between contamination and treatment. Moreover, the discrepancy between our results and those of Ryabova (10) could have been attributed to some differences in experimental design, such as the route of radiocaesium administration. It seems that a satisfactory explanation remains to be elucidated in further experiments.

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