

IAEA-TECDOC-926

***The use of Prussian Blue to reduce  
radiocaesium contamination of  
milk and meat produced on  
territories affected by  
the Chernobyl accident***

*Report of United Nations Project E 11*



INTERNATIONAL ATOMIC ENERGY AGENCY

IAEA

February 1997

The IAEA does not normally maintain stocks of reports in this series.  
However, microfiche copies of these reports can be obtained from

INIS Clearinghouse  
International Atomic Energy Agency  
Wagramerstrasse 5  
P.O. Box 100  
A-1400 Vienna, Austria

Orders should be accompanied by prepayment of Austrian Schillings 100,—  
in the form of a cheque or in the form of IAEA microfiche service coupons  
which may be ordered separately from the INIS Clearinghouse.

The originating section of this publication in the IAEA was:

FAO/IAEA Agriculture and Biotechnology Laboratory  
Agency's Laboratories  
International Atomic Energy Agency  
Wagramerstrasse 5  
P.O. Box 100  
A-1400 Vienna, Austria

**THE USE OF PRUSSIAN BLUE TO REDUCE RADIOCAESIUM CONTAMINATION OF  
MILK AND MEAT PRODUCED ON TERRITORIES AFFECTED BY  
THE CHERNOBYL ACCIDENT**

**IAEA, VIENNA, 1997**

**IAEA-TECDOC-926**

**ISSN 1011-4289**

**© IAEA, 1997**

**Printed by the IAEA in Austria  
February 1997**

## FOREWORD

The accident at the Chernobyl nuclear power plant in 1986 resulted in the contamination of large tracts of agricultural land and forests in northern Europe but particularly in Belarus, the Russian Federation and the Ukraine. Of particular radiological significance still now are  $^{137}\text{Cs}$  and  $^{90}\text{Sr}$  which migrate through the soil-plant-animal food chain and accumulate in milk and meat consumed by the human population inhabiting these contaminated regions.

The implementation of certain land reclamation and organizational measures has made it possible to maintain much of the milk and meat produced within the temporary permissible levels (TPLs) for radionuclides adopted by different States. The most reliable and widely used method of reducing the contamination of livestock products after surface contamination with radionuclides has occurred is to feed uncontaminated forage or forage with a low radionuclide content. This is achieved through a variety of measures including: fencing off contaminated land, so prohibiting access to livestock; restricting livestock to indoor maintenance and provision with purchased 'clean' feeds and forage; improving access to freshly sown pasture and excluding forage harvested from natural and unimproved meadows; application of potassium fertilizers and lime to reduce radionuclide uptake by crops; and substitution of contaminated pasture with maize and other cereals, silage, fodder beet and hay produced on arable land. These countermeasures, although extremely costly, remain very effective in reducing the radionuclide levels in meat and milk produced by State and collective farms. However, for very many small farmers in CIS countries the use of these countermeasures is hampered by cost, elevated rates of transfer of radiocaesium from soil into plants, and by the inaccessibility of the meadows commonly grazed such as in forest clearings or in marshy areas to any agromechanical countermeasures. Where 'traditional' countermeasures have not been able to be employed and where  $^{137}\text{Cs}$  and  $^{90}\text{Sr}$  contamination have been considered to be too high, whole communities (hamlets, villages, towns) have been translocated to 'clean' locations; this has had a very negative psychosocial impact on the well-being of the population.

However, it was thought possible that the radiocaesium content of milk and meat could be reduced by the simple administration to livestock of materials that bind radiocaesium in the gastrointestinal tract and thereby decrease its absorption and increase its excretion in faeces. Prussian Blue is the name given to a number of ferric hexacyanoferrates which have these binding properties. When introduced into the gastrointestinal tract, it slowly dissolves into a soluble colloidal form where it reacts with dietary radiocaesium to form a complex that cannot penetrate biological membranes.

Within the framework of a collaborative project investigations were conducted between 1990 and 1995 to evaluate the use in cattle of Prussian Blue compounds (in the form of boli, salt licks, or direct addition to the diet) for reducing the radiocaesium content of milk and meat, and the subsequent effect of dung from treated animals on the transfer of radiocaesium from soil to plants. In association with these studies, investigations were conducted to monitor the possible effects of Prussian Blue administration on milk and meat composition and the physiological well-being of cattle. The possible toxicological effects of feeding rats with milk and meat from animals treated with Prussian Blue was also investigated.

This report summarizes the results of the laboratory studies and field trials carried out in the CIS. However, in essence, it represents the cumulative effort of over one hundred scientific and collaborative staff in seven countries over a six year period. The report is composed of ten sections in which an attempt has been made to synthesize into a relatively few pages the immense amount of data collected; much of this is reported in the annexes, especially Annex B. The body of the report not only deals with technical aspects of the use of Prussian Blue in livestock, but also with its economic, agricultural, radiological, toxicological and socio/psychological considerations in human populations who have to cope with contamination due to 'fallout'.

It is concluded that Prussian Blue materials are simple to use, extremely cost-effective, and significantly reduce radiocaesium levels in the meat and milk of cattle grazing contaminated land. As

a consequence of these studies, the feeding or application of Prussian Blue was officially approved by the Ministries of Agriculture of Belarus, the Russian Federation and the Ukraine, and particularly recommended for use by the small livestock farmer.

This report is the outcome of a collaborative project between the Food and Agriculture Organization of the United Nations, the International Atomic Energy Agency, the Agricultural University and Radiation Hygiene Institute of Norway, the Ukrainian Research Institute of Agricultural Radiology (Kiev), the Byelorussian Institute of Agricultural Radiology (Gomel), the Russian Institute of Agricultural Radiology and Radioecology (Obninsk), the Queens University (Belfast, United Kingdom). Financial support was provided by the Governments of Norway and the three CIS countries affected by the accident.

### *EDITORIAL NOTE*

*In preparing this publication for press, staff of the IAEA have made up the pages from the original manuscript(s). The views expressed do not necessarily reflect those of the governments of the nominating Member States or of the nominating organizations.*

*Throughout the text names of Member States are retained as they were when the text was compiled.*

*The use of particular designations of countries or territories does not imply any judgement by the publisher, the IAEA, as to the legal status of such countries or territories, of their authorities and institutions or of the delimitation of their boundaries.*

*The mention of names of specific companies or products (whether or not indicated as registered) does not imply any intention to infringe proprietary rights, nor should it be construed as an endorsement or recommendation on the part of the IAEA.*

## CONTENTS

|  |    |
|--|----|
| 1. BACKGROUND TO THE PROBLEM . . . . .   | 7  |
| 1.1. Overview of possible countermeasures . . . . .  | 7  |
| 1.2. Milk production, marketing and monitoring . . . . .   | 7  |
| 1.3. Beef production, marketing and monitoring . . . . .   | 8  |
| 2. INTERNATIONAL AND STATE INTERVENTION LEVELS FOR RADIOACTIVE<br>CONTAMINATION OF MILK AND MEAT . . . . . | 8  |
| 3. BINDING OF CAESIUM IONS BY PRUSSIAN BLUE . . . . .  | 10 |
| 4. DOSES OF PRUSSIAN BLUE COMPOUNDS REQUIRED FOR<br>GRAZING ANIMALS . . . . .                              | 11 |
| 5. ADMINISTRATION OF PRUSSIAN BLUE TO FARM ANIMALS . . . . .   | 11 |
| 5.1. Direct addition to feed or compounding into concentrate . . . . .                                     | 11 |
| 5.2. Salt licks . . . . .  | 12 |
| 5.3. Sustained release boli . . . . .  | 12 |
| 6. TOXICOLOGICAL ASPECTS . . . . .   | 12 |
| 7. SUMMARY OF EXPERIMENTS CARRIED OUT IN BELARUS,<br>THE RUSSIAN FEDERATION AND THE UKRAINE . . . . .      | 13 |
| 8. UTILIZATION OF PB COMPOUNDS IN THE CIS . . . . .  | 16 |
| 9. THE BENEFITS OF USING CAESIUM BINDERS . . . . .   | 16 |
| 9.1. Dose reductions achievable . . . . .  | 16 |
| 9.2. Saving other resources . . . . .  | 16 |
| 9.3. Sociological factors and psychological considerations . . . . .                                       | 19 |
| 10. CONCLUSIONS AND RECOMMENDATIONS . . . . .  | 19 |
| ANNEX A: BOLUS COMPOSITION AND TREATMENT . . . . .   | 23 |
| ANNEX B: FIELD EXPERIMENTS PERFORMED IN THE THREE REPUBLICS . . . . .                                      | 27 |
| Studies performed in the Russian Federation . . . . .  | 27 |
| Studies performed in the Ukraine . . . . .   | 43 |
| Studies performed in Belarus . . . . .   | 56 |
| ANNEX C: TOXICOLOGICAL CONSIDERATIONS . . . . .  | 69 |
| BIBLIOGRAPHY . . . . .   | 75 |
| CONTRIBUTORS TO DRAFTING AND REVIEW . . . . .  | 79 |
| FAO/IAEA PUBLICATIONS ON AGRICULTURAL AND OTHER CONSEQUENCES<br>OF THE CHERNOBYL ACCIDENT . . . . .        | 80 |

# 1. BACKGROUND TO THE PROBLEM

## 1.1. OVERVIEW OF POSSIBLE COUNTERMEASURES

The accident at the Chernobyl nuclear power plant in April 1986 led to the dispersal of radioactive material over many countries. In the initial period after the accident, problems were related to iodine contamination; however, since then the predominant problems have been associated with radiocaesium ( $^{137}\text{Cs}$ ) and radiostrontium ( $^{90}\text{Sr}$ ) contamination. Because of their geographic situation, the countries most affected by fallout from the accident have been Belarus, the Russian Federation and the Ukraine. In each of these States there are two principal livestock production systems - dairy and beef - and these are practiced on both collective and private farms (Tables 1 and 2). Since the Chernobyl accident, many different countermeasures have been employed in the affected areas to ensure that foodstuffs produced for human consumption have radiocaesium levels below legal levels set by each state (so-called "temporary permissible levels" (TPLs)). For example, pastures on state farms have been 'decontaminated' by deep ploughing and re-seeding the land, or through the application of lime and potassium fertilizers. Typical three-fold reductions in radiocaesium transfer have been obtained by ploughing procedures. These improvements have led to a situation where radiocaesium levels in milk and meat produced by the collective state farms are generally lower than the TPLs. Nevertheless in 1992, approximately 100 000 dairy cows in Belarus and the Ukraine, many kept by small-scale private farmers to provide milk for their families and local communities, still produced milk that exceeded the adopted national control levels for radiocaesium of 111 Bq/L and 370 Bq/L, respectively. In the Russian Federation, a further 15 000 animals were reported to produce milk with levels above their adopted TPL of 370 Bq/L.

## 1.2. MILK PRODUCTION, MARKETING AND MONITORING

In all three States, the sale, processing and/or consumption of milk that exceeds these TPLs is illegal. Consequently, all contaminated milk *should* be used either directly for animal feeding or exchanged with the state/local authority for clean milk and used subsequently by them for making butter or animal feed. Although there are no data available, in reality a not insignificant proportion of this contaminated milk is actually consumed by the small farmer and his family or the rural community in general. There is no price differential between clean and contaminated milk.

TABLE I. ESTIMATE OF DAIRY CATTLE NUMBERS AND MILK PRODUCTION IN THE CONTAMINATED AREAS OF BELARUS, UKRAINE AND THE RUSSIAN FEDERATION, 1992-1994

|                           | Total No. dairy cattle in contaminated area | Average milk production/ animal (litres/annum) | Temporary Permissible Level, TPL Cs-137 (Bq/L) | No. cows producing milk above TPL |
|---------------------------|---|--|--|-----------------------------------|
| <b>BELARUS</b>            |   |  |  |                                   |
| Collective                | 393 000                                     | 3 000  | 185 <sup>a</sup>                               | 3 000                             |
| Private                   | 84 000                                      | 3 600  | 185  | 25-30 000                         |
| <b>UKRAINE</b>            |   |  |  |                                   |
| Collective                | 170 000                                     | 2 500  | 370  | 43 000                            |
| Private                   | 60 000                                      | 3 500  | 370  |                                   |
| <b>RUSSIAN FEDERATION</b> |   |  |  |                                   |
| Collective                | 200 000                                     | 2 700  | 370  | 7 000                             |
| Private                   | 25 000                                      | 3 000  | 370  | 3 000                             |

<sup>a</sup> TPL reduced to 111 Bq/L in 1993

TABLE II. ESTIMATE OF NUMBERS OF CATTLE USED FOR BEEF AND NUMBER OF ANIMALS EXCEEDING INTERVENTION LEVELS IN BELARUS, UKRAINE AND THE RUSSIAN FEDERATION, 1992-1994

|                    | Total No. beef cattle in contaminated area | Live weight at slaughter (kg) | Temporary Permissible Level, TPL Cs-137 (Bq/kg) | No. cattle above TPL after 3 months on 'clean' feed |
|--------------------|--|-------------------------------|---|---|
| Belarus            | 700 000                                    | 420                           | 600   | 4 400   |
| Ukraine            | 500 000                                    | 400 - 420                     | 740   | 3 000   |
| Russian Federation | 300 000                                    | 400 - 450                     | 740   | 5 000   |

Currently all milk from collective State farms is monitored on entry into the milk factory. Any milk not meeting the appropriate standards is made into butter or sent for animal feed. Milk from animals on private farms is also monitored by local food hygiene authorities (under the Ministry of Health). Samples are taken twice monthly from 10% of animals in a given village; not less than ten samples are collected from each village. Farmers are informed of the results, and they receive privileges when the milk is contaminated above the TPL; all contaminated milk is bought by the State.

### 1.3. BEEF PRODUCTION, MARKETING AND MONITORING

The systems of marketing animals for meat are similar throughout the areas under consideration. Animals from collective and private farms are purchased by the State/local authority and sent either to local abattoirs for slaughter or to fattening centres. As a rule, no distinction is made between the two types of production systems. Beef production and marketing are much less affected by contamination than milk production since, prior to slaughter, animals are generally fed on 'clean' forage/grain for 2-3 months by which time levels of radiocaesium are generally below the TPLs. Nevertheless, even under this system, in 1992 more than 10 000 animals/year exceeded the TPLs of 600 Bq/kg (in Belarus) and 740 Bq/kg (in Ukraine and the Russian Federation), Table II. Therefore, these animals had to be maintained on expensive clean feeds for a considerable time periods (1-3 months) before slaughter.

Whilst the practice of feeding clean feed and monitoring animals and their products now used in the three States guarantees the production of meat that complies with the TPLs for radiocaesium, the severe shortages of concentrate feeds (estimated at 4 million tons in Belarus alone) and the foreseen privatization of land will bring about rapid changes in the pattern of agriculture, including beef production. This will put a severe strain on the ability of the authorities to ensure that all meat available to the consumer is below the stated TPLs. The dramatic decrease in grain and clean herbage production from the contaminated zones for feeding animals during the finishing period has significantly reduced the use of these areas for beef production.

## 2. INTERNATIONAL AND STATE INTERVENTION LEVELS FOR RADIOACTIVE CONTAMINATION OF MILK AND MEAT

In the years following the accident many national authorities and international organizations addressed the problem of contamination of food with radionuclides by setting so-called intervention levels. The Codex Alimentarius Commission (a joint body of FAO and WHO), which proposes acceptable standards for additives and contaminants in foodstuffs moving in international trade, adopted levels for radiocaesium of 1000 Bq/kg for both milk and meat. In the European Union the present radiocaesium limit for milk and dairy produce is 370 Bq/L while for meat it is 600 Bq/kg.



The state intervention levels (or Temporary Permissible Levels - TPLs) for milk and meat respectively in the three CIS countries in 1994 were: Belarus (111 Bq/L and 600 Bq/kg), Ukraine (370 Bq/L and 740 Bq/kg), and Russia (370 Bq/L and 600 Bq/kg). However, these limits are frequently lower at the regional level, e.g. in the Gomel region in Belarus, the TPLs for <sup>137</sup>Cs in milk and meat were 37 Bq/L and 370 Bq/kg at the time this report was compiled.

TABLE III. GUIDELINE LEVELS FOR RADIONUCLIDES IN FOODS FOLLOWING ACCIDENTAL NUCLEAR CONTAMINATION FOR USE IN INTERNATIONAL TRADE

CAC/GL 5-1989

| Dose per unit intake factor (Sv/Bq)    | Representative radionuclides                           | Level (Bq/kg) |
|--|--|---------------|
| FOODS DESTINED FOR GENERAL CONSUMPTION |  |               |
| 10 <sup>-6</sup>                       | <sup>241</sup> Am, <sup>239</sup> Pu                   | 10            |
| 10 <sup>-7</sup>                       | <sup>90</sup> Sr                                       | 100           |
| 10 <sup>-8</sup>                       | <sup>131</sup> I, <sup>134</sup> Cs, <sup>137</sup> Cs | 1000          |
| MILK AND INFANT FOODS                  |  |               |
| 10 <sup>-5</sup>                       | <sup>241</sup> Am, <sup>239</sup> Pu                   | 1             |
| 10 <sup>-7</sup>                       | <sup>131</sup> I, <sup>90</sup> Sr                     | 100           |
| 10 <sup>-8</sup>                       | <sup>134</sup> Cs, <sup>137</sup> Cs                   | 1000          |

**Notes:** These levels are designed to be applied only to radionuclides contaminating food moving in international trade following an accident and not to naturally occurring radionuclides which have always been present in the diet. The Codex Alimentarius Guideline Levels remain applicable for one year following a nuclear accident. By an accident is meant a situation where the uncontrolled release of radionuclides to the environment results in the contamination of food offered in international trade.

As the proposed levels have extensive conservative assumptions built in, there is no need to add contributions between dose per unit intake groups, and each of the three groups should be treated independently. However, the activity of the accidentally contaminating radionuclides within a dose per unit intake group should be added together if more than one radionuclide is present. Thus the 1000 Bq/kg level for the 10<sup>-8</sup> Sv/Bq dose per unit intake group is the total of all contaminants assigned to that group. For example, following a power reactor accident, <sup>134</sup>Cs and <sup>137</sup>Cs could be contaminants of food, and the 1000 Bq/kg refers to the summed activity of both these radionuclides.

These levels are intended to be applied to food prepared for consumption. They would be unnecessarily restrictive if applied to dried or concentrated food prior to dilution or reconstitution.

The levels apply to situations where alternative food supplies are readily available. Where food supplies are scarce, higher levels can apply.

Both FAO and WHO have called attention in the expert meeting reports to special consideration which might apply to certain classes of food which are consumed in small quantities, such as spices. Some of the foods grown in the areas affected by the Chernobyl accident fall-out contained very high levels of radionuclides following the accident. Because they represent a very small percentage of total diets and hence would be very small additions to the total dose, application of the Guideline Levels to products of this type may be unnecessarily restrictive. FAO and WHO are aware that policies vary at present in different countries regarding such classes of food.

TABLE IV. SPECIFIC INTERVENTION LEVELS (OR TEMPORARY PERMISSIBLE LEVELS) CURRENTLY EMPLOYED IN CONTAMINATED AREAS<sup>a</sup> OF CIS COUNTRIES (1995) FOR WITHHOLDING FOODS FROM HUMAN CONSUMPTION

|                    | <sup>137</sup> Cs in milk<br>Bq/L | <sup>137</sup> Cs in meat<br>Bq/kg |
|--------------------|-----------------------------------|------------------------------------|
| Belarus            | 111 <sup>b</sup>                  | 600 <sup>b</sup>                   |
| Ukraine            | 370                               | 740                                |
| Russian Federation | 370                               | 740                                |

<sup>a</sup> Defined as areas where deposition of <sup>137</sup>Cs, <sup>90</sup>Sr and <sup>239</sup>Pu fallout still exceed 1, 0.15 and 0.01 Ci/km<sup>2</sup> (or 37, 5, 0.4 kBq/m<sup>2</sup>) respectively.

<sup>b</sup> Some contaminated districts use even lower levels (e.g. Gomel region uses 37 Bq/L milk and 370 Bq/kg meat).

The International Chernobyl Project concluded that the intervention levels operating in the three States in 1990 were unnecessarily restrictive from a radiologic standpoint and that there was a case for relaxation of the levels to at least those of the Codex Alimentarius Commission.

### 3. BINDING OF CAESIUM IONS BY PRUSSIAN BLUE

Following the Chernobyl accident investigations were carried out in a number of countries including Austria, France, Germany, Norway and the United Kingdom on the use of Prussian Blue (PB) for reducing radiocaesium levels in a wide range of animals (cattle, sheep, pigs, goats, reindeer, deer, poultry) and their products.

PB compounds are coloured complexes consisting of a hexacyano-moiety and polyvalent cations like iron, nickel, copper or cobalt. All of these compounds bind caesium ions in exchange for a monovalent cation. There are a range of different PB compounds that reduce radiocaesium contamination. The five substances tested in the present project were:

- Ammonium ferric cyanoferrate (AFCF)  $\text{NH}_4\text{Fe}[\text{Fe}(\text{CN})_6]$
- Potassium ferric cyanoferrate (KFCF)  $\text{K Fe}[\text{Fe}(\text{CN})_6]$
- Ferric cyanoferrate (FCF)  $\text{Fe}_4[\text{Fe}(\text{CN})_6]_3$
- Ferrocyn Mixture of 95% FCF and 5% KFCF
- Bifege Mixture of 10% Ferrocyn and 90% Cellulose.

These compounds have been used for producing salt licks, boli and concentrates and for addition to roughages. The caesium-binding properties exhibited by the compounds are explained by the physico-chemical properties of the crystal unit cells which form cubicles centered around an exchangeable cation (sodium, potassium or ammonium). The PB compounds form colloids in aqueous media, and the particle size of the colloids (5-50  $\mu\text{m}$ ) is important in providing a large surface area for the binding of caesium. At equal molar concentrations, caesium is bound  $10^3$  to  $10^4$  times more efficiently to the hexacyanoferrate complex than sodium or potassium.

Binding of caesium occurs within the physiological pH range of the gut, and in fluids with ionic compositions that can be expected during digestion in animals and man. Colloidal particles of PB compounds are too large to be absorbed. Caesium bound to PB is therefore not available for absorption, but is excreted in the dung. There it remains bound to PB for an extended period of time. (Experiments carried out during the project showed that caesium bound to PB is taken up at a reduced rate by plant roots.)

Prussian Blue compounds act primarily by reducing the rate of gastrointestinal absorption of radiocaesium, and therefore basically have the same effect as feeding clean feeds. As an additional, minor contribution to the reduction in radiocaesium content of contaminated animals, PB compounds bind some of the radiocaesium from the body pools recycled to the gastrointestinal tract (endogenous secretion) and prevent its reabsorption. To maintain the effect, it is necessary to have a continuous presence of the compound in the digestive tract.

The optimal techniques for administration will therefore differ greatly depending on the agricultural production systems in question, and the availability on a daily basis of concentrates, other supplementary feeds or cut forage. In meat producing animals PB is only required during the last 1-2 months before slaughter while in dairy animals PB must be provided on a daily basis either orally or from a depot (a bolus) within the digestive tract.

Shortly after the Chernobyl accident, PB compounds were used in animals in Scandinavia for reducing the radiocaesium content of their products; and following the radiological accident in Goiânia, Brazil in 1987, PB was also effectively used in man to reduce the burden of radiocaesium in the body.

#### **4. DOSES OF PRUSSIAN BLUE COMPOUNDS REQUIRED FOR GRAZING ANIMALS**

PB compounds are the most effective caesium binders available on a unit weight basis. Dose levels recommended vary in the literature, but are generally 3 g/day for cows (6 mg/kg body weight) and 1-2 g/day (10-40 mg/kg body weight) for smaller ruminants. At these dose levels, reduction factors of up to 10-fold (90% reduction in transfer) or more have been reported both in milk and meat. Experiments on the effects of lower daily doses of PB show a dose dependent reduction in caesium binding activity with a 2-fold reduction factor at doses of as low as 0.8 - 1 mg/kg body weight of Giese salt (AFCF with 36%  $\text{NH}_4\text{Cl}$ ). Other PB compounds, including Ferrocyn produced in Ukraine, KFCF from Belarus and KCoHCF, are all active at low concentrations, albeit with about half the efficiency of Giese salt. Their high efficiency makes PB compounds particularly useful for radiocaesium decontamination in grazing ruminants. In the present situation, daily doses of as low as 1 mg/kg body weight will result in sufficient reductions in caesium levels (50-60%) in animals producing milk and meat with contamination levels above the present TPLs. Although higher doses will reduce radiocaesium transfer by 90% or more, this rate of administration may be difficult to achieve in grazing animals which are not fed or handled on a daily basis.

#### **5. ADMINISTRATION OF PRUSSIAN BLUE TO FARM ANIMALS**

There are various methods of administering PB: a) in concentrate feed, particularly useful in intensive production systems; b) by sprinkling directly on to forage, a cheaper but more labour intensive alternative; c) in salt licks and blocks, and d) as boli, especially well suited for grazing animals or when no concentrate is offered.

##### **5.1. DIRECT ADDITION TO FEED OR COMPOUNDING INTO CONCENTRATE**

On small farms with a few animals, PB can be conveniently given as a powder added to a portion of grain, concentrate or roughage. Ferrocyn and Bifege have been successfully administered in this form to cattle in the CIS. In 1993 and 1994 packages of ferrocyn, providing a daily dose of 2-3 g per cow, were distributed to village farmers in contaminated areas in the Ukraine. In Russia, Bifege has been regarded as a more convenient method for administration since the powder can be distributed in bulk and scooped into the feed in larger amounts than pure PB. About 6 000 animals were fed Bifege in 1994.

When industrial feed compounding facilities are available, the easiest and cheapest method of feeding PB is by its direct incorporation into concentrate feed. In this way, it is not necessary to give any special instruction to the farmer on how to use the PB. Experimental studies have demonstrated the effectiveness of incorporating PB into concentrate rations provided to ruminants. In Norway, this procedure has been used since 1988 in the form of a special concentrate containing 1 g/kg of PB. Since livestock need to receive the concentrate every day the procedure is primarily suitable for intensive production systems or for 'hand-fed' animals. In the CIS, PB is added to farm compounded concentrate (cereal) mixes.

## 5.2. SALT LICKS

If concentrate is not available, or where it is not feasible to offer concentrate because animals are grazing extensively over longer periods of time, salt licks can be used. Since 1989, about 100 tons of salt lick containing 2.5% PB have been used annually by more than 300 000 sheep in Norway. The lick must have sufficient physical strength to withstand the inclement weather conditions in the mountains for the full length of a grazing season. In areas where the sodium content of the vegetation is low, sheep and cattle are accustomed to visiting salt licks. Since 1989, PB salt licks have been employed in Norwegian mountain pastures and a 2-fold reduction in radiocaesium content has been achieved each grazing season. An average daily intake of 1 g salt (25 mg/day of PB) gives a 50% reduction in radiocaesium levels in a 30-40 kg lamb; and 10 g/day gives a similar reduction in a fattening steer. The efficiency of the salt lick depends upon its constant access in order to ensure frequent consumption (daily or several times per week). However, in a grazing herd there are always some animals who are not interested in salt. The use of AFCF salt licks must therefore be accompanied by some monitoring of milk and meat for  $^{137}\text{Cs}$ . In the CIS, PB salt licks have been increasingly employed since 1990 and are officially approved for use. Although this avenue of administering PB only reduces  $^{137}\text{Cs}$  levels in milk and meat by up to two-fold, nevertheless it is the easiest method of administration by far. In Belarus, the authorities employed 300 000 salt blocks in the Gomel region in 1993; the licks weighed 5 kg and contained 6% PB in the form of ferrocyn. In 1994, 33 tons of PB salt licks were used for dairy and beef cattle. Salt licks have not been used so extensively in Ukraine. In the Russian Federation, salt briquettes (containing PB) are now commonly employed on collective farms in contaminated regions.

## 5.3. SUSTAINED RELEASE BOLI

The low daily doses of PB required make caesium binders well suited for administration by slow release boli placed in the rumen. In large scale field experiments in Norway, treatment with sustained release boli reduced the radiocaesium levels of whole herds; the frequency of failures was below 1% when live monitoring was performed 6 weeks after treatment. In these studies, boli with 15-20% AFCF remained in the rumen for at least 2 months. The release of AFCF during this period reduced radiocaesium accumulation in meat by 50-80% over a 6-8 week period. Boli are now authorized for routine use in cattle, sheep and reindeer in Norway; boli weighing 200 g have been tested in dairy and beef cattle. The results are similar to those obtained in sheep, with reductions in milk radiocaesium levels dependent on the release rate of PB. At least two boli have to be given in order to achieve adequate release rates. The composition of the boli used in Norway and in the experiments in Belarus, the Russian Federation and Ukraine was similar. Figure 1 illustrates the effect of bolus administration in a dairy cow consuming contaminated pasture.

## 6. TOXICOLOGICAL ASPECTS

Studies on possible metabolic and toxicological effects of PB compounds date back to the early 1960's. These investigated the use of PB for reducing the radiocaesium content of agricultural products and for reducing the internal dose of individuals contaminated accidentally. Experiments were carried out on laboratory rodents, domestic animals and in man. In addition to its use as a caesium binder, PB is also recommended as a therapeutic antidote for thallium poisoning.

Furthermore, it is pertinent to note that ferrocyanide compounds are permitted as food additives, e.g. as anticaking agents in table salt.

Despite these conclusions, the CIS scientists involved in this project wanted to further confirm the findings and broaden the number of parameters investigated.

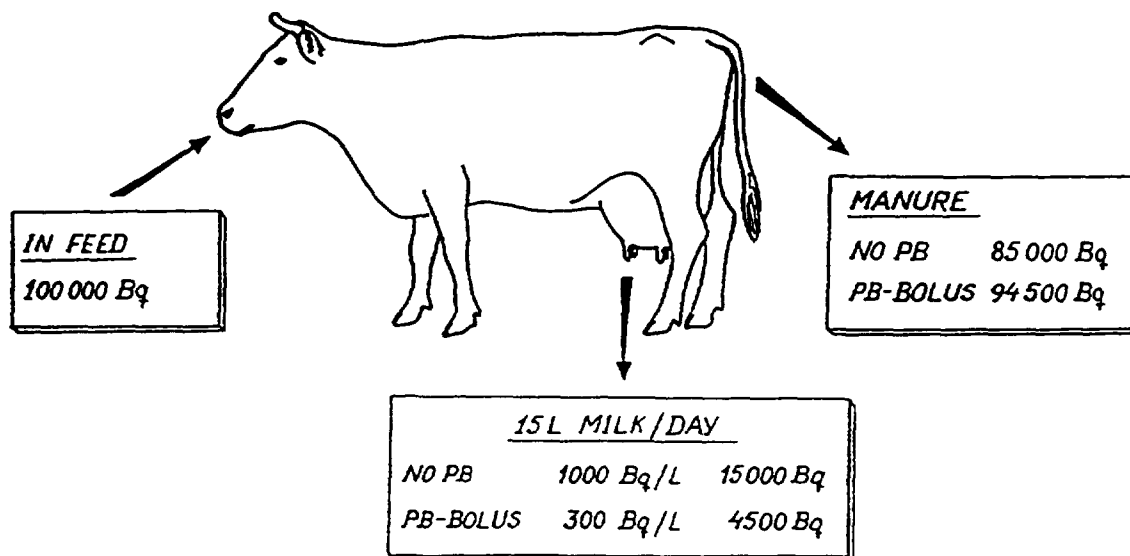


FIG. 1. Effect of PB-bolus on the transfer of radiocaesium (Bq/d) to milk. Comparison of the radiocaesium budget in a cow with PB boli and an untreated cow. Constant radiocaesium levels in milk and manure would be obtained after several weeks of grazing on a contaminated pasture.

The conclusions from these studies are fully described in Annex C of this report. In summary, it can be concluded that PB compounds:

- have no adverse effects on animal health and production;
- have no toxic effects in man when used experimentally or therapeutically;
- the consumption of milk and meat from PB treated animals is not expected to have any negative effect on human health.

## 7. SUMMARY OF EXPERIMENTS CARRIED OUT IN BELARUS, THE RUSSIAN FEDERATION AND THE UKRAINE

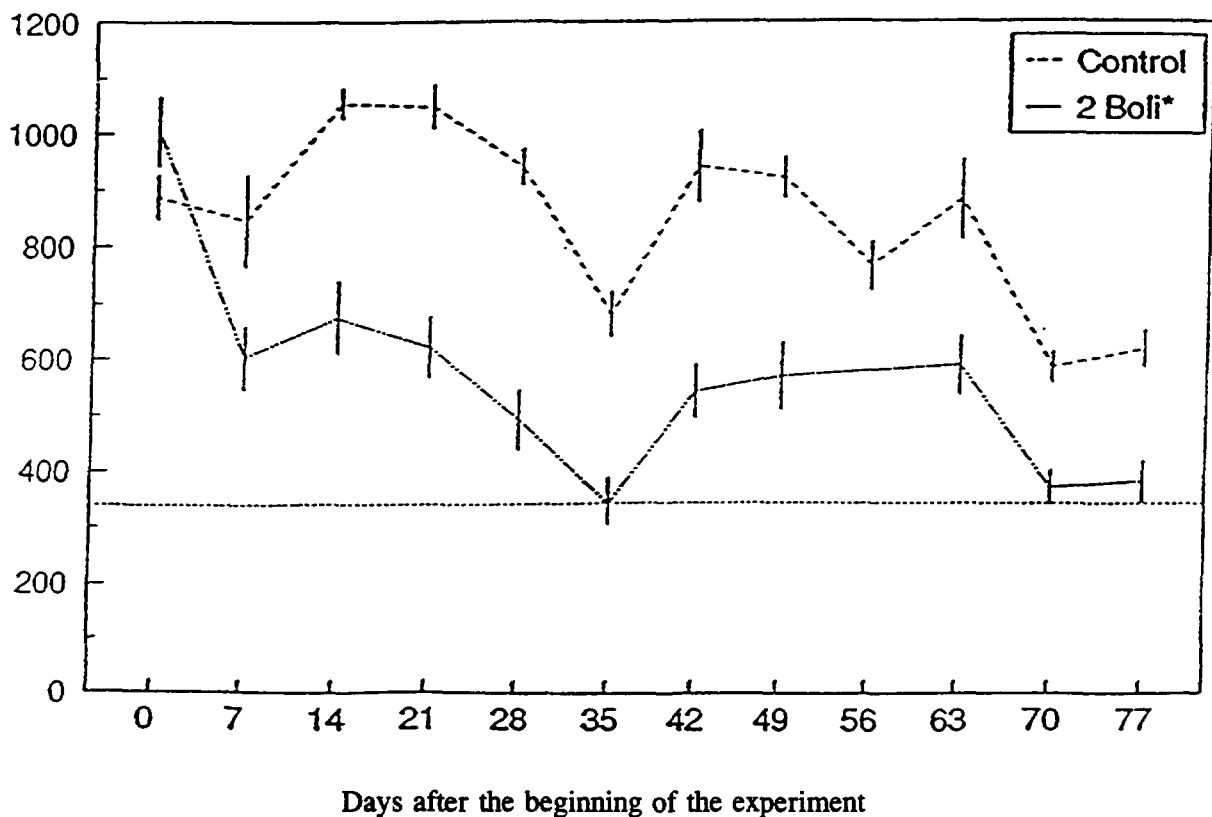
● As part of the International Chernobyl Project, initial experiments which began at the end of the 1990 grazing season showed that the administration of PB boli was potentially suitable for field application in the contaminated areas of Belarus, the Russian Federation and Ukraine. Field trials were subsequently undertaken during the summer months of 1991-1994 to obtain practical results from the use of this technology in reducing radiocaesium concentrations in milk and meat from farm animals grazing contaminated pastures, and to further investigate possible toxicological or biological problems that might arise.

● Studies conducted in the Ukraine on dairy cows showed 2-4 fold reductions in the radiocaesium content of milk during the main part of the grazing season following treatment with 2 or 3 boli in May and 2 or 3 more boli in July 1991 (Figures 2 and 3).

- It was agreed that a practical schedule for treatment involving administration of  $3 \times 200$  g boli each cow in May, July and August/September would guarantee a reduction in radiocaesium levels in the milk of all treated animals by a factor of 3-5. This would allow cows to produce 'clean' milk while grazing pastures that, under normal circumstances, would produce milk with a radiocaesium level of approximately 1100-1200 Bq/L.

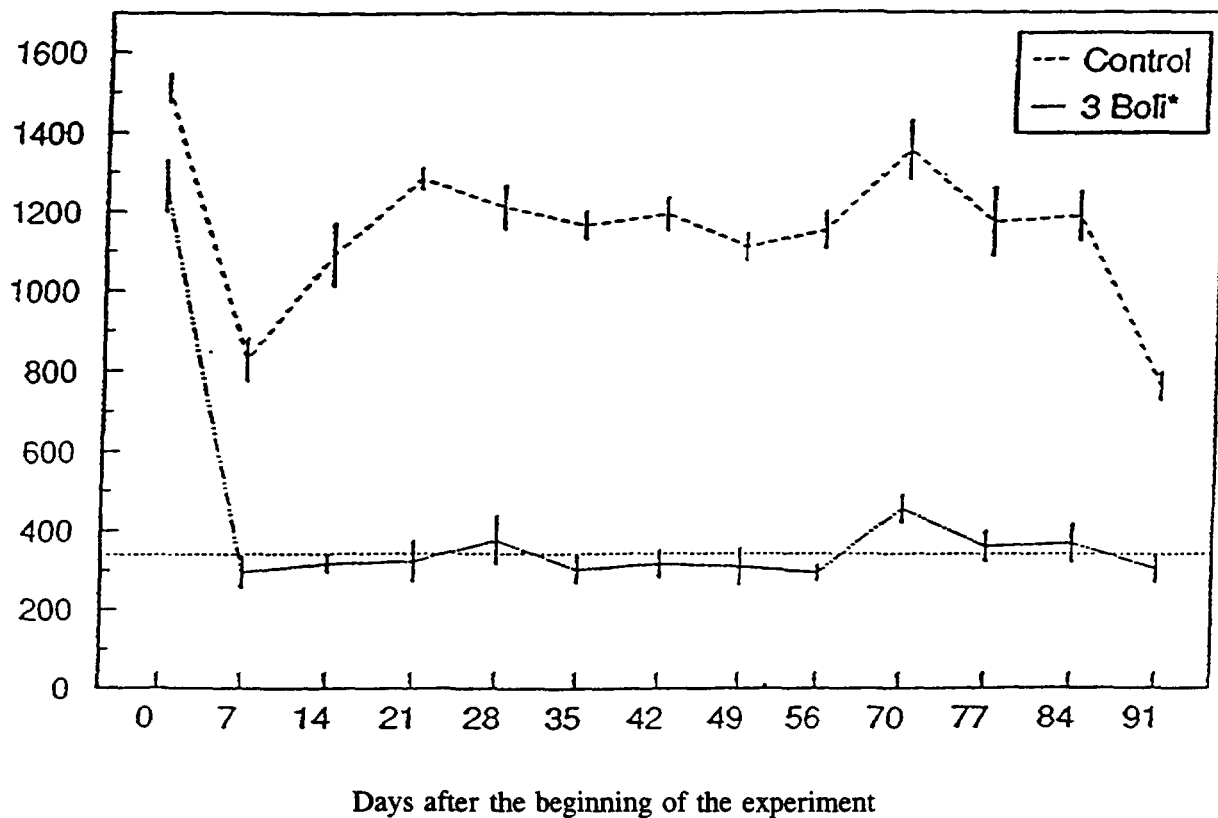
- In beef cattle, treatment with  $2 \times 200$  g boli led to a reduction of radiocaesium levels in meat by a factor of 3.5 when they were slaughtered 90 days after treatment. Somewhat higher reductions were seen after 60 days due to the higher release rate of PB from the boli at that time. Therefore, in beef cattle, treatment with boli combined with grazing of moderately contaminated pastures is an effective alternative to the commonly used indoor feeding of finishing rations during the last 2-3 months before slaughter. Bolus techniques also have potential in the proposed utilization of contaminated areas for meat production.

- A large set of parameters relevant to the health and nutritional status of treated animals was examined in these experiments. No signs of macro- or trace-element deficiencies or toxicities attributable to the bolus treatment were found. Haematological parameters, body weight gain and milk production were similar to those recorded in untreated control animals.



\* 2 boli were introduced per os at day 0 and day 60

FIG. 2. <sup>137</sup>Cs concentration in the milk (Bq/kg) of experimental cows in private farms of village Lugovoye in Ukraine. 10 cows per group. Average and standard deviations. Boli were given on day 0 and on day 60.



\* 3 boli were introduced per os at day 0 and day 60

FIG. 3. <sup>137</sup>Cs concentration in the milk (Bq/kg) of experimental cows in private farms of village Velikiye Ozera in Ukraine. 15 animals in each group. Average and standard deviations. Boli were given on day 0 and day 60.

- Dung from bolus-treated animals was tested in field experiments as a fertilizer for sandy soils on which oats and lupins were being grown. The Prussian Blue from the boli reduced the content of radiocaesium in plants by a factor of two during the first growing season; however, during the second season no significant reduction in radiocaesium levels could be detected. Further studies need be carried out to clarify the benefits of applying dung from PB bolus treated livestock on soils where vegetables are grown for human consumption.

- Using PB synthesized in the Ukraine, boli were manufactured at the Agricultural University of Norway according to the methods described in Annex A. Variations in the composition of the wax in the boli were tested in a separate set of experiments. However, the substitution of the original beeswax by wax mixtures more readily available in the CIS was not successful since release rate of PB by some formulations was too slow, while other formulations were not physically stable in the rumen. It is therefore recommended that boli contain the following: 15-25% PB, 60-75% barite mineral and 10-15% bees-wax as outlined in Annex A.

## **8. UTILIZATION OF PB COMPOUNDS IN THE CIS**

In the Ukraine field studies were carried out in 20 villages in the Rovno Region in 1993 and in 70 villages in both the Zhitomir and Rovno Regions in 1994. A total of 6 500 dairy cattle were given both boli and powdered ferrocyn. In confirmation of the previous experimental studies ferrocyn reduced the radiocaesium content of milk by factors of 2 to 4.5. Ferrocyn containing boli reduced radiocaesium levels in meat 3-6 fold. In 1995, the Government budgeted on US \$40 000 (equivalent to 2-3 tons) for the purchase of ferrocyn from Russia. However, demand for PB is estimated to be 6 tons.

In Belarus, approximately 27 tons of PB was used per year to treat 30 000 cows during the three year period, 1993-1995. In 1993 and 1994, 9 000 and 16 000 cows respectively were given PB boli; in addition, 33 tons of PB salt licks were provided in 1994. In 1995, 4000 tons of PB containing concentrate were produced.

## **9. THE BENEFITS OF USING CAESIUM BINDERS**

It has been shown above that caesium binders reduce the levels of contamination in meat and milk, and in crops fertilized with the resulting manure. The practical benefits that would ensue by using the technique fall into three main categories: reduction in radiological dose, saving of resources and psycho-social benefits.

### **9.1. DOSE REDUCTIONS ACHIEVABLE**

In theory, if the concentration of radiocaesium in meat and milk is reduced by a factor of three, then so too is the radiological dose to people consuming this food. Assuming that 15% of the annual dose is currently due to exposure to external irradiation and 85% to internal irradiation, and that 90% of the internal dose is from consumed milk and meat, one can calculate that reducing the radiocaesium content of milk and meat by a factor of three would imply an overall reduction in dose of about 60%. For territories where there is a high uptake of caesium from soil to pasture, this overall dose reduction would be higher (probably greater than 80%).

Unfortunately, the situation is more complicated than this. Whereas it is illegal to market meat or milk with levels of radiocaesium above the TPLs in order to protect the human population from exposure to high internal doses, it has been reported that, especially under the present economic conditions, many subsistence farmers use the milk from their animals for consumption in their own households even when the milk has levels in excess of the TPLs. The use of PB compounds would lead to immediate reductions in radiation dose to these people of the order of 60% or more as outlined above.

The collective dose saved by implementing the technique is not easy to estimate. However, if it assumed that there is an average 3-fold reduction in the radiocaesium concentration of milk consumed (i.e. to below the TPL) and that some 50 million litres of milk are involved, a very approximate figure for the collective dose saved is of the order of 200 man·Sv. This modest saving is a direct result of using extremely low TPLs and does not detract from the significant cost-benefit ratio of the use of PB compounds.

### **9.2. SAVING OTHER RESOURCES**

The use of Prussian Blue through the medium of boli, salt licks or direct addition to the diet can save significant resources currently being expended on other methods of reducing caesium contamination in milk and meat. The cost effectiveness of using caesium binders will depend upon prevailing levels of pasture contamination, the costs of production and distribution of the binders and upon the cost of alternative strategies. However, with the current economic difficulties in the three



States, the lack of true market prices reflecting the inherent value of commodities, the huge devaluation of the Rouble currencies and high inflation rates, it is extremely difficult to quantify the benefits in Rouble terms. In order to present the potential savings, it has been necessary to give a perspective based on Western experience where market prices more accurately reflect value, and also to quantify the savings in physical terms such as numbers of animals, litres of milk and hectares of land affected.

Table V illustrates typical levels of <sup>137</sup>Cs in milk and meat from cows grazing pasture with levels of <sup>137</sup>Cs contamination ranging from 200-10 000 Bq/kg herbage. These figures assume a daily pasture intake of 70 kg by a 500 kg cow (i.e. 14% of body weight). Also illustrated are <sup>137</sup>Cs levels when Prussian Blue is administered according to the recommendations given earlier. These data clearly demonstrate that by administering Prussian Blue by the most suitable methods, both milk and meat can be produced that satisfy the presently adopted TPLs even in animals grazing pasture contaminated by up to 1500 Bq/kg herbage (i.e. herbage typical of a large part of the land area presently contaminated in the three States).

TABLE V. RELATIONSHIP BETWEEN LEVELS OF PASTURE CONTAMINATION AND Cs-137 LEVELS IN MEAT AND MILK, AND THE EFFECT OF ADMINISTERING BOLI ON MEAT AND MILK LEVELS

| Level of pasture contamination (Bq/kg) | Intake/day <sup>a</sup> (Kbq) | Meat                             |   | Milk                            |  |
|--|-------------------------------|----------------------------------|---|---------------------------------|--|
|  |                               | Cs-137 level equilibrium (Bq/kg) | Cs-137 level following boli treatment (Bq/kg) | Cs-137 level equilibrium (Bq/L) | Cs-137 level following boli treatment (Bq/L) |
| 250                                    | 17.5                          | 280                              | 90  | 112                             | 34   |
| 500                                    | 35                            | 700                              | 234   | 280                             | 94   |
| 1 000                                  | 70                            | 1 400                            | 460   | 560                             | 186  |
| 1 500                                  | 105                           | 2 100                            | 700   | 840                             | 280  |
| 2 000                                  | 140                           | 2 800                            | 920   | 1 120                           | 374  |
| 3 000                                  | 210                           | 4 200                            | 1 400   | 1 680                           | 560  |
| 5 000                                  | 350                           | 7 000                            | 3 000   | 2 800                           | 920  |
| 10 000                                 | 700                           | 14 000                           | 4 600   | 5 600                           | 1 860  |

<sup>a</sup> Assumes daily intake of 70 kg fresh herbage/animal

Based on the data presented in Table V, a cost-benefit ratio between PB boli treatment per annum and the value of additional 'clean' milk which would become available as a result can be calculated. In Belarus, KFCF can currently be imported from Germany at a cost of about US \$7/kg and from the plant in Kursk (Russian Federation) for about US \$20/kg. With a maximum of 9 boli used for a 6-7 month grazing season, 300 g of PB at a price of US \$2.50 will be required. (In 1995, the real cost of producing 9 boli was 45 000 Belarus Roubles.) Assuming that the costs of treatment and transportation amounts to the same as the production costs, the additional cost of producing around 1 800 litres of milk suitable for human consumption (assuming a daily production of 10 litres/cow/day over the 6 month grazing season) is 90 000 Belarus Roubles. This milk would have a market value of approximately 3.6 million Roubles (2 000 R/litre milk). Thus, the use of PB boli would provide a cost:benefit ratio of 1:40. However, if the saving on radiological dose to a population is sufficient to avoid translocation, an extremely unpopular and expensive countermeasure, the cost:benefit ratio of PB could be 10-fold greater, viz. > 1:400.

Using the data in Table I on the number of animals presently producing milk above TPLs it may be calculated that by making PB boli available to private farmers, an additional 350 million litres of 'clean' milk could be provided annually for local product manufacture or liquid consumption in Belarus, the Russian Federation and the Ukraine at extremely low cost. The Treasuries of the three States would also benefit in that they would no longer have to buy contaminated milk from the farmers at considerable cost and the pressure to compensate the people which live on contaminated territories would be reduced.

Each of the three States has already made huge investments in the provision of relatively clean herbage by ploughing, draining, fertilizing, liming and reseeding natural pastures on state and collective farms. These efforts have undoubtedly succeeded in reducing contamination in herbage and hence milk and meat to within the TPLs, and will also serve to improve animal productivity over the long term. It is not clear to what extent this process is complete, but it has been variously reported to be 80-90%. The remaining land (10-20%) is largely that owned by small private farmers and of such small size, poor quality or accessibility that traditional land decontamination procedures are uneconomic, difficult or inappropriate. The provision of PB compounds to cattle would allow this land to be used for meat production, since animals could be taken directly from pasture to slaughter. In areas where levels of <sup>137</sup>Cs in meat remain above TPLs in spite of PB treatment, the period required for feeding clean feeds could be shortened by approximately 40-50 days, thus saving a considerable amount of harvested feed.

The cost of producing and transporting clean feed for animals in the private sector and of transporting finished animals to 'clean' feed centres is not possible to quantify with accuracy, but it may be estimated that today in Belarus and the Ukraine, 35 000 - 40 000 animals (requiring 70 000 - 80 000 hectares of additional clean pasture) are subjected to this procedure annually prior to slaughter. The area required could be reduced to between 15 000 and 18 000 hectares if PB were used. Utilizing current EU estimates, providing this additional improved pasture land would cost US \$400/ha when spread over 10 years (this is the expected period over which <sup>137</sup>Cs pasture levels will remain unacceptably high assuming no countermeasures are taken together with an annual maintenance cost of US \$100/hectare), it can be calculated that the meat would cost an additional US \$1/kg to produce in order to satisfy existing international levels in each of the three States over this period. The alternative, i.e. administering PB materials for up to three months prior to slaughter, would cost an additional US 10 cents/kg, viz. approximately ten times less.

When considering these cost-benefit ratios and how they might differ from Western countries, several observations can be made. Firstly, PB is readily available in the CIS although beeswax or a suitable substitute for producing boli is sometimes difficult to obtain. Secondly, the labour required for the manufacture and distribution of PB materials would be relatively cheap in the three States and the costs of transportation and distribution would be far less than the high cost of regularly collecting 'dirty' milk and distributing 'clean' feed. On balance it may therefore be concluded that the cost:benefit ratios calculated provide adequate demonstration of the economic benefits of the technique both for CIS and Western European countries.

It should also be recognized that with the economic and political changes now taking place in the CIS, there is the potential for large growth in the private farming sector, at least in the long term. Inevitably this will mean that farmers themselves (and not the authorities) will need to have methods available to reduce the radiocaesium content in milk and meat. The PB technique (bolus, concentrate or salt lick) is eminently suited for this new development; it might also be appropriate as a means of addressing other deficiencies in animal nutrition through the provision of supplementary microelements.

Finally, at present, there are laws in the three States that define compensation payments to people whose total annual radiological dose exceeds a specific value. By using PB compounds, it may be estimated that the number of people who would receive such compensation would fall by about one-half. For Belarus alone, this could mean approximately 100 000 people being brought out from this scheme with a consequent saving of a few percent of the national budget.

### 9.3. SOCIOLOGICAL FACTORS AND PSYCHOLOGICAL CONSIDERATIONS

During the International Chernobyl Project [see Bibliography] some sociological/psychological factors were identified that amplified feelings of concern among the exposed populations in the contaminated territories. These were often related to people's feeling of impotence - they perceived no means of personally controlling or alleviating the hazards. Numerous studies have shown that stress depends crucially upon the way the particular stressor is perceived by the individual and his/her assessment of personal and external resources for controlling it. Many psychological disturbances can result from this stress and it has been recognized that prolonged experience of failure to control environmental changes leads to a state of chronic apathy.

In the contaminated areas there is clearly anxiety about the levels of caesium contamination in the environment and fears about the possible presence of other less publicized radionuclides, particularly strontium and plutonium. There is still uncertainty about the long term health consequences of the high levels of exposure in the first few days and weeks following the accident.

Some of the countermeasures currently used, such as prohibiting subsistence farmers from consuming milk and meat produced from their own animals, have considerable negative psychological, social and financial consequences.

During the field trials carried out as part of the work, it was clear to the scientists involved in the project that the farming community welcomed the opportunity to use the caesium binders. Indeed there was considerable dissatisfaction from the owners of the "control groups" of animals that they were not allowed the opportunity of using the binders. Whilst this reaction has not been analysed by social scientists, it is clear that the widespread use of PB compounds in the contaminated areas would allow farming practices to return to normal, with a resultant improvement in the quality of life of very many farming families. Whilst there existed some understandable reaction against the use of 'chemicals' by a few, the "fear" of radiation and the disturbing influence of alternative countermeasures on people's lives were of more importance to the vast majority of those affected. However, it is imperative that information on the caesium binder technique, the scientific/ technical basis for its effectiveness, its non-toxic nature, the procedure for its administration to livestock and the availability of local monitoring support should be made available to the public, before and during implementation. Activity levels in milk and meat should also be made available, and information presented on how these compare with country standards, EU standards and the recommendations of the FAO/WHO Codex Alimentarius Commission.

## 10. CONCLUSIONS AND RECOMMENDATIONS

Laboratory experiments and field trials conducted on collective and private farms during 1990-1994 in Belarus, the Russian Federation and the Ukraine showed that the application of PB compounds was highly efficient in reducing radiocaesium concentrations in milk and meat in animals grazing contaminated pasture. Results from toxicological studies showed no deleterious effects. A series of physiological and biochemical tests on treated animals showed no deviations from the values obtained in control animals. This is in agreement with all the reports cited in the international scientific literature.

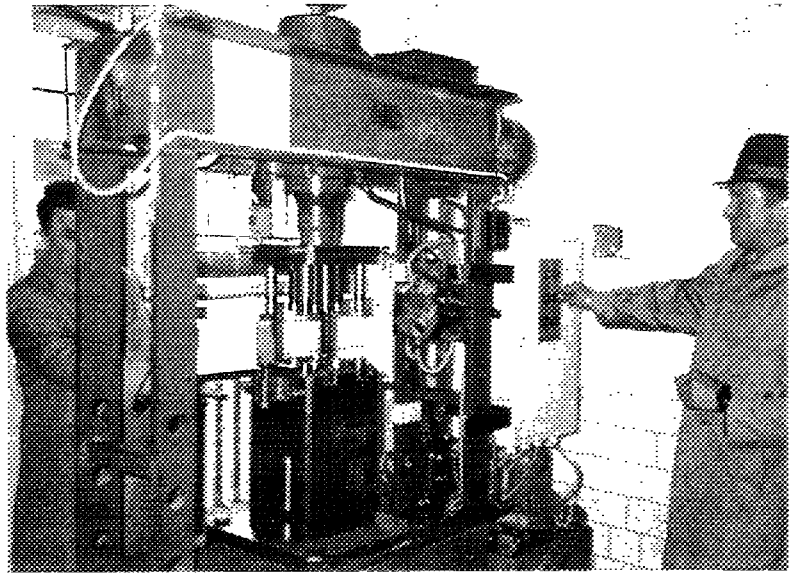
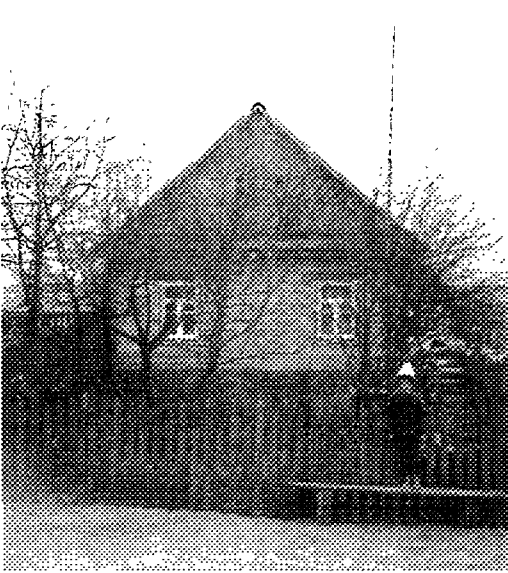
The benefits of utilizing PB compounds are considerable and are summarized in Table VI.

TABLE VI. SUMMARY OF THE BENEFITS OF USING PRUSSIAN BLUE COMPOUNDS

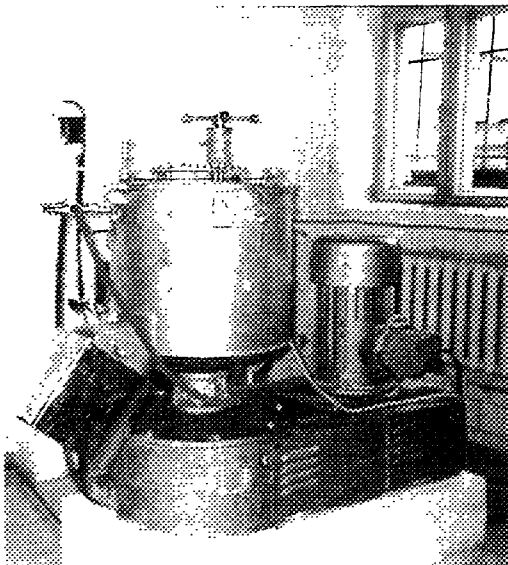
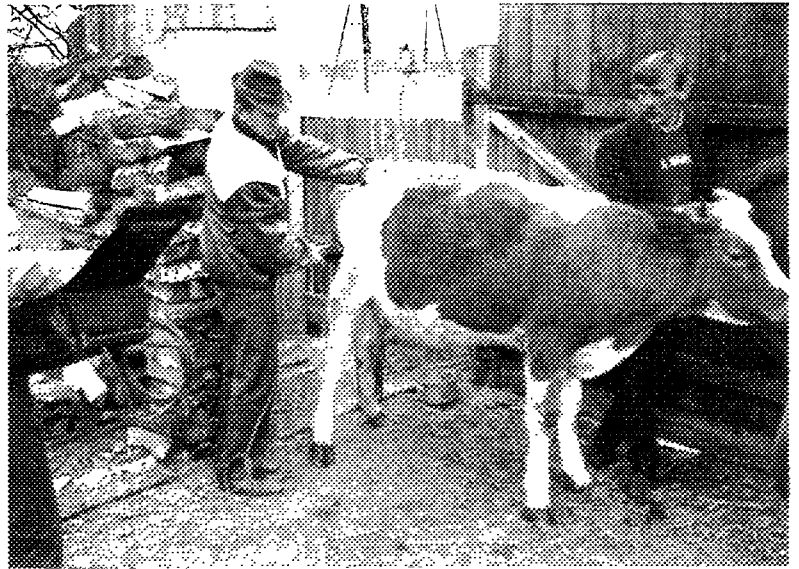
| Benefit                               | Comment  |
|---------------------------------------|--|
| Individual dose reduction             | Overall reduction of about 60% (probably more than 80% in areas where the uptake factor from soil to grass is particularly high).  |
| Collective dose                       | Perhaps a few hundred man·Sv; relatively small because of the extremely low TPLs being used in the States. Nevertheless it is cost-effective.  |
| Additional milk production            | An additional 50 million litres of milk per annum would meet TPLs without the need for distribution of "clean" feed and milk.  |
| Clean feed needed for meat production | The time needed for "clean feeding" could be reduced by 40 - 50 days, resulting in a reduction by a factor of 5 of the area of improved pasture needed.  |
| Social/psychological                  | Some 50 000 farmers could return to traditional farming practices with a corresponding increase in their sense of well-being and improvement in their quality of life. Many farmers destined for translocation would no longer have to move house. |
| Compensation                          | The number of individuals receiving compensation for exceeding an annual dose criterion could be reduced by approximately 50%.   |

As a result of the experiments and field trials described in this report:

- **Prussian Blue was licensed in the CIS as an animal feed additive for administration in concentrate, salt lick or sustained release boli.**
- **Permission was given for the sustained release bolus technique to be implemented on a large scale.**
- **Adequate funds were made available in all three States to give sufficient technical and economic support for the utilization of PB compounds for reducing radiocaesium content in milk and meat.**
- **The application of PB compounds is well suited for the future infrastructure of dairy and beef production in the CIS. The potential additional benefits of using boli to deliver other compounds that would increase the productivity of livestock (e.g. minerals, anthelmintics) or to carry potential strontium binders in the CIS is recognized.**
- **Studies should continue on the long-term binding of radiocaesium to PB in soil and the impact on pasture and field crops.**



In southern Belarus, where areas were affected by radioactive fallout from the Chernobyl accident, small farmholders are applying countermeasures to reduce levels of contamination in milk, meat, and other products. They are being assisted through projects supported by the Norwegian government and the FAO/IAEA programme. Shown here is a typical small farm in the region; equipment for mixing "Prussian Blue" compounds and for making boli which are used to reduce radiocaesium levels in cows; and scientists monitoring the body gamma radiation of livestock given Prussian Blue. (Credit: Richard IAEA)



**NEXT PAGE(S)  
left BLANK**

## ANNEX A

### BOLUS COMPOSITION AND TREATMENT

#### Composition of boli

Sustained release boli remain in the rumen for a prolonged period (many weeks) and release active ingredients into the rumen liquor. Typically, a bolus is made up of the following material (w/w):

---

|               |     |          |
|---------------|-----|----------|
| Prussian Blue | 20% | (15-25%) |
| Beeswax       | 15% | (10-15%) |
| Barite        | 65% | (60-75%) |

---

The values in brackets may be used to modify the efficiency of the bolus, but it should be noted that the density of a bolus should be above  $2.0 \text{ g cm}^3$  in order to prevent regurgitation. Barite is a mineral (primarily a barium sulphate complex) commonly used as heavy ballast in the oil industry; in a bolus, it is finely ground ( $<0.1 \text{ mm}$ ). The ingredients are mixed together at  $80^\circ\text{C}$  and then spread out to solidify. The material can then be ground to beads of less than 1 g, or made into boli by compression in stainless steel moulds using a pressure of  $3\text{-}5 \times 10^8 \text{ Pascal}$ . For cattle, boli are ideally 35 mm in diameter and 200 g weight.

No standardized equipment for bolus production is available on the market. In Norway conventional hydraulic presses have been used to operate the piston in the cylinder which makes the boli. Control of the movements of the piston by a PLS-system which facilitates programming of the different phases of the piston movement, has enabled 1-2 boli per minute to be produced with the pilot equipment developed. In Belarus, technology is in place to produce 4000 boli per day; and many thousands of boli are being manufactured both in the Ukraine and in Russia. The chemicals used in the production of boli are non-toxic although care should be taken in handling PB, which, as a strongly coloured compound, may discolour both machines and their surroundings.

#### Shortage of wax

A shortage and the high price of beeswax have been noted as possible constraints to the production of boli in the three States. Therefore joint experiments have been carried out with waxes and fats that are more readily available. Boli were made in Kiev and tested both in animals that received contaminated feeds in Ukraine, and in fistulated cows, which enabled the observation of the rate of dissolution by weighing the boli at regular intervals after insertion.

Two types of wax formulations were tested:

|         |           |     |                  |                |                    |
|---------|-----------|-----|------------------|----------------|--------------------|
| Type 1: | Barite    | 68% | Wax composition: | Paraffin       | 55%                |
|         | Ferrocyne | 20% |                  | Cerezine       | 45%                |
|         | Wax       | 12% |                  |                |                    |
| Type 2: | Barite    | 68% | Composition CKF: | Paraffin       | 55%                |
|         | Ferrocyne | 20% |                  | Cerezine       | 35%                |
|         | Alloy CKF | 12% |                  | Butylcautchouk | 5%                 |
|         |           |     |                  | Vaseline oil   | 5% (medical grade) |

Type 1 boli dissolved too slowly; the rate of release of PB gave rise to radiocaesium reductions in milk of less than 10%.

Boli made from the type 2 formulation were not stable in the rumen, and disintegrated within 2 to 4 weeks of insertion.

Fistulated cow studies carried out in May-September 1991 showed that the mean release rates of PB from boli made from beeswax (3 boli/cow) were:

| Time after start of experiment (weeks) | 0-2   | 2-4 | 4-6  | 6-8  | 8-10 | 8-10 |
|--|-------|-----|------|------|------|------|
| Ferrocene release rate (mg/d)          | 1 455 | 800 | 645  | 635  | 435  | 794  |
| Ferrocene release rate (% available/d) | 1.2   | 0.8 | 0.73 | 0.80 | 0.61 | 1.2  |

Future studies may provide wax mixes with the same reproducible release rates as the beeswax mixes. Until such time, it is recommended that beeswax formulations are used for bolus production.

### Availability of Prussian Blue

The cheapest source of Prussian Blue is in the Russian Federation; there is none commercially available in Belarus and the Ukraine. Import-export and exchange control regulations between the countries comprising the CIS can lead to delays in the delivery of Prussian Blue to Belarus and the Ukraine; consequently, considerable forward-planning is required to ensure adequate supplies are available for manufacture into boli at the start of a grazing season.

### Treatment of animals with PB boli

The bolus has a dome-shaped front end which aids in swallowing. The head of the animal is held by one person and the bolus is inserted into the mouth by means of a standard applicator. In order to facilitate swallowing, the bolus must be placed in the pharynx, well behind the back of the tongue. Animals normally swallow the bolus immediately, and with the boli used in Ukraine, Belarus and the Russian Federation no loss of boli was observed during the treatment period. Several thousand animals have been treated with boli in the CIS. Out of approximately 31 000 animals treated in Belarus, four animals died due to asphyxiation from a bolus being inserted into the trachea rather than the oesophagus. These cows were particularly nervous; it is therefore recommended that boli should not be administered in such cases.

Several hundred animals including cattle, sheep, goats and reindeer have been examined after slaughter for injuries to the stomach epithelium or other internal complications relating to the presence of boli at the bottom of their reticulo-rumens without finding any evidence of pathological changes.

### Production of salt licks

Prussian Blue salt licks are produced by the same technology as that used for producing ordinary sodium chloride licks. Prussian blue is mixed with salt in a 2.5% (w/w) concentration before compression into 10 kg blocks in a hydraulic press. Higher concentrations of PB may be included (5-10%) although care should be taken to ensure that the physical strength of the block is not reduced significantly as a result of the anticaking properties of PB compounds.

### **Combined use of PB in boli and salt licks**

It is well reported in the literature that the effect of PB in reducing radiocaesium absorption is dependent on the concentration of PB in the rumen and the nature of the intestinal contents. Therefore one might expect additive effects of the simultaneous use of both boli and salt licks. Salt licks reduce the average radiocaesium level in a flock or herd and are very useful for grazing animals because the labour needed for administration is nil. However, salt licks carry an uncertainty when the radiocaesium levels of individual animals are concerned, since some animals may purposely or otherwise avoid the lick and thus have negligible PB intakes. Thus, boli provide a more guaranteed reduction of radiocaesium levels in milk and meat from every animal in the treated herd whereas salt licks are strongly recommended as a way of reducing radiocaesium levels of the average animal. A combination of salt licks and boli may reduce radiocaesium levels by a factor of 4-7 (2-5 fold reduction for the bolus and 2-fold reduction for the salt lick). The more highly contaminated animals may be identified by live monitoring, isolated from the herd and offered either clean feed and pasture and/or bolus treatment.



## ANNEX B

### FIELD EXPERIMENTS PERFORMED IN THE THREE REPUBLICS

#### Introduction

Most of the scientific work described in this paper was carried out following discussions and agreements reached at a meeting convened in Oslo (12-15 March 1991) on the use of PB for decreasing the radiocaesium content of animal products (meat, milk); the agreed protocol was dated 15 March 1991.

The objectives of the collaboration between the IAEA, FAO, the CIS, Agricultural University of Norway and the Norwegian Radiation Protection Authority were as follows:

- to carry out practical trials on the use of PB boli under normal milk/beef production conditions on both State farms and private small holdings;
- to carry out comparative tests of the efficiency of locally produced PB and that manufactured in Western Europe.

#### STUDIES PERFORMED IN THE RUSSIAN FEDERATION

All the research and experimental work was organized and conducted by the Research Institute of Agricultural Radiology, Obninsk. Laboratory studies in animals on the effectiveness and possible harmful effects of ferrocyanide began in 1989. Field trials were conducted between 1990 and 1994. In 1991 an experiment of 4 months duration on 18 cows (10 given PB boli and 8 controls) was carried out in the Novozybkov district of Bryansk region, at the Novozybkov Branch of the Institute of Fertilizers and Agricultural Soil Science (NBIFASS). The experimental work was aimed at:

- measuring the efficiency of PB boli in reducing radiocaesium contamination;
- monitoring the effect of administering a second batch of boli two months after the first;
- estimating any possible effects on soil-plant transfer of radiocaesium fixed Prussian Blue in dung.

Between 1992 and 1994 trials were conducted on four collective farms. The aim was to further test the effects of Prussian Blue under practical conditions in the collective farming system. (All cows owned by small farmers had been taken away in 1986.)

#### Materials and methods

In 1991 due to the strict demands placed by the Regional Veterinary Service on the experimental conditions (see the experimental method), the number of cows was limited to a random division of 10 'treated' and 8 'controls'.

The animal groups were formed on 3 May 1991, and feeding of forage with increased levels of radiocaesium was started immediately (the rations had a variable caesium content of  $9-20 \times 10^4$  Bq/kg; total daily radiocaesium intake amounted to ca.  $2 \times 10^5$  Bq). The first 3 PB boli were given by Norwegian specialists with the participation of Russian counterparts. After one month confinement in a stable, the cows were put out to pasture together with the main herd. A second application of 3 boli was given by Russian staff, without assistance, on 19 June 1991. The experiments were completed on 20 September 1991.

The following measurements were recorded for all cows:

- General physiological status (body temperature, pulse, respiration) and behaviour were noted daily when feeding.
- Number and quality of rumen movements were estimated by palpation.
- Estimation of the erythrocyte, leucocyte and thrombocyte levels in blood.
- Estimation of the plasma thyroid hormone content by radioimmunoassay (RIA).
- Estimation of blood thiocyanate content using the method of Degiampietro *et al.* (1987).
- Measurement of micro- and macro-elements in plasma by atomic absorption analysis.
- Milk yields were recorded in individual control milkings.
- Radiocaesium concentrations in milk samples were measured using gamma-spectrometry.
- Radiocaesium measurements on components of the ration were also periodically performed.

## RESULTS OF STUDIES CONDUCTED IN 1991 AND 1992

As Table B.1. shows, radiocaesium levels in milk of cows given PB boli was 1.5 - 5 times lower than that of the control group. Radiocaesium concentrations in the milk of untreated animals ranged from  $1.0 \times 10^5$  Bq/L at the beginning of the experiment, when hay with high  $^{137}\text{Cs}$  content ( $1.4 \times 10^5$  Bq/kg) was deliberately introduced into the ration, to 77 Bq/L when on pasture. Treated animals were kept under the same conditions and fed the same pastures throughout (Tables B.2. and B.3.). The radiocaesium content in the cows' diet during the period ranged from  $1 - 3 \times 10^4$  Bq, depending on the regimes and period of grazing on the pastures mentioned above. Whatever the total radiocaesium content in the diet, the effect of the boli in reducing the radionuclide content in milk amounted to 1.5 to 4-fold.

It should be noted that the radiocaesium content of milk samples from individual cows varied from day to day (Table B.4.). For example, milk from cow No. 0701 one month after the first administration of boli (5.06) had a radiocaesium level of 370 - 430 Bq/L; at the same time the untreated cow No. 3141, from the control group, had almost the same level of radiocaesium. Such phenomena may be found in individual cows on all dates of the experiments.

### The clinical assessment of PB application to livestock

A number of clinico-physiological and biochemical investigations were carried out to demonstrate that the boli did not affect the health and well-being of the animals. No deviations from normal were observed during the study. The animals were healthy and milk yields in the control and treated animals were the same. Table B.5. shows the average daily milk yields. The data show that there were no differences in productivity between the control and experimental animals throughout the investigation. There were no differences in blood parameters between the control and experimental cows (Table B.6.).

The investigation of hormones of the thyroid gland (Table B.7.) showed a decrease in the levels of thyroxine ( $T_4$ ) and triiodothyronine ( $T_3$ ) in blood. However, low levels of thyroid hormones are typical of all cattle in this zone due to the endemically low iodine content of the soil.

When applying ferrocyanide-containing preparations, the presence of cyanide groups is of profound concern. Therefore, a single animal blood analysis was carried out at the end of the experiment, i.e. after 4 months of the permanent presence of ferrocyanide in the gastrointestinal tract. The data obtained are given in Table B.8. and these show that there were no differences in the thiocyanate content of the blood of experimental and control animals.

In view of the prospect of using sorbents to reduce the uptake and accumulation of radiocaesium in milk and meat, some authors have suggested the possibility of combining these sorbents with limiting essential trace- and macro-elements. The blood of all the experimental animals

was analysed for 12 trace- and macro-elements (Tables B.9. and B.10.). The results did not show any differences between treated and untreated animals for any of these indices.

Thus, the administration of ferrocyanide-containing boli produces a reduction in the transfer of radiocaesium to milk by a factor of 1.5 - 4, depending on the time after the administration of boli. The duration of the effect of the boli is about 2 months. No negative effects on the health of the cows were revealed from the detailed clinico-physiological or biochemical indices investigated.

TABLE B.1. Cs-137 ACTIVITY IN COWS MILK, NBIFASS FARM, BRYANSK REGION

| Date  | Untreated group   |  | Treated group     |  |
|-------|-------------------|--|-------------------|--|
|       | Number of animals | Cs-137 concentration Bq/L ( $\times 10^{-8}$ Ci/L) | Number of animals | Cs-137 concentration Bq/L ( $\times 10^{-8}$ Ci/L) |
| 11-05 | 8                 | 992 $\pm$ 85 (2.68 $\pm$ 0.23)                     | 10                | 936 $\pm$ 41 (2.53 $\pm$ 0.11)                     |
| 15-05 | 8                 | 925 $\pm$ 74 (2.50 $\pm$ 0.20)                     | 10                | 288 $\pm$ 15 (0.78 $\pm$ 0.04)                     |
| 22-05 | 8                 | 300 $\pm$ 30 (0.81 $\pm$ 0.08)                     | 10                | 118 $\pm$ 7 (0.32 $\pm$ 0.02)                      |
| 29-05 | 8                 | 862 $\pm$ 59 (2.33 $\pm$ 0.16)                     | 10                | 222 $\pm$ 37 (0.60 $\pm$ 0.10)                     |
| 05-06 | 8                 | 618 $\pm$ 56 (1.67 $\pm$ 0.15)                     | 10                | 185 $\pm$ 30 (0.50 $\pm$ 0.08)                     |
| 12-06 | 8                 | 655 $\pm$ 52 (1.77 $\pm$ 0.14)                     | 10                | 241 $\pm$ 33 (0.65 $\pm$ 0.09)                     |
| 20-06 | 8                 | 292 $\pm$ 18 (0.79 $\pm$ 0.05)                     | 10                | 100 $\pm$ 11 (0.28 $\pm$ 0.03)                     |
| 27-06 | 8                 | 170 $\pm$ 7 (0.46 $\pm$ 0.02)                      | 10                | 81 $\pm$ 15 (0.22 $\pm$ 0.04)                      |
| 04-07 | 8                 | 152 $\pm$ 11 (0.41 $\pm$ 0.03)                     | 9                 | 89 $\pm$ 11 (0.24 $\pm$ 0.03)                      |
| 19-07 | 8                 | 329 $\pm$ 41 (0.89 $\pm$ 0.11)                     | 6                 | 204 $\pm$ 44 (0.55 $\pm$ 0.12)                     |
| 29-07 | 8                 | 126 $\pm$ 15 (0.34 $\pm$ 0.04)                     | 6                 | 33 $\pm$ 7 (0.09 $\pm$ 0.02)                       |
| 20-08 | 8                 | 78 $\pm$ 7 (0.21 $\pm$ 0.02)                       | 6                 | 26 $\pm$ 11 (0.07 $\pm$ 0.03)                      |
| 29-08 | 8                 | 303 $\pm$ 18 (0.82 $\pm$ 0.05)                     | 5                 | 67 $\pm$ 15 (0.18 $\pm$ 0.04)                      |
| 20-09 | 8                 | 89 $\pm$ 11 (0.24 $\pm$ 0.03)                      | 5                 | 48 $\pm$ 18 (0.13 $\pm$ 0.05)                      |

First bolus application - 3 May 1991

Second bolus application - 19 June 1991

TABLE B.2. RADIOCAESIUM ACTIVITY IN PASTURE GRASS AT NOVOZYBKOV INSTITUTE

| Sample N | Pasture type                          | Area, hectares | Cs-137 concentration |                    |
|----------|---------------------------------------|----------------|----------------------|--------------------|
|          |                                       |                | Bq/kg                | ( $10^{-8}$ Ci/kg) |
| 1        | Natural unimproved green bulk         | 4              | 536                  | (1.45)             |
| 2        | Natural unimproved green bulk         |                | 115                  | (0.31)             |
| 3        | Natural unimproved green bulk         | 5              | 5 590                | (15.10)            |
| 4        | Natural unimproved green bulk         | 20             | 218                  | (0.59)             |
| 5        | Complementary seeding without backset |                | 111                  | (0.3)              |
| 6        | Natural                               | 2.5            | 207                  | (0.56)             |
| 7        | Natural                               | 12             | 93                   | (0.25)             |
| 8        | Sown grass                            | 12             | 19                   | (0.05)             |
| 9        | Natural                               | 8              | 3 050                | (8.25)             |
| 10       | Radical improvement                   | 18             | 59                   | (0.16)             |
| 11       | Natural                               | 6              | 1 206                | (3.26)             |
| 12       | Natural                               | 6              | 96                   | (0.26)             |

TABLE B.3. Cs-137 ACTIVITY IN COW RATION

| Before 91-05-03<br>(started giving contaminated hay) |         | K Bq/kg | (Ci/kg)                   |
|--|---------|---------|---------------------------|
| Sown-grass hay                                       | 5 kg    | 20.3    | ( $0.55 \times 10^{-6}$ ) |
| Silage   | 15 kg   | 0.37    | ( $0.01 \times 10^{-6}$ ) |
| Barley meal  | 3 kg    | 0.37    | ( $0.01 \times 10^{-6}$ ) |
| TOTAL:   | 23 kg   | 21.1    | ( $0.57 \times 10^{-6}$ ) |
| From 91-05-03  |         |         |                           |
| Natural meadow hay                                   | 1.5 kg  | 203     | ( $5.5 \times 10^{-6}$ )  |
| Silage   | 15 kg   | 0.37    | ( $0.01 \times 10^{-6}$ ) |
| Barley meal  | 3 kg    | 0.37    | ( $0.01 \times 10^{-6}$ ) |
| TOTAL:   | 19.5 kg | 203     | ( $5.5 \times 10^{-6}$ )  |
| From 91-06-13<br>(after the experiment was finished) |         |         |                           |
| Pasture grass  | 40 kg   | 14.8    | ( $0.4 \times 10^{-6}$ )  |
| Green feed   | 20 kg   | < 0.1   | ( $0.1 \times 10^{-8}$ )  |
| Barley meal  | 3 kg    | < 0.1   | ( $0.1 \times 10^{-8}$ )  |
| TOTAL:   | 63 kg   | 14.8    | ( $0.4 \times 10^{-6}$ )  |

TABLE B.4. RADIOCAESIUM CONCENTRATIONS IN COW'S MILK ( $\times 10^{-8}$  Ci/L) TREATED OR UNTREATED WITH PB BOLI AT THE RUSSIAN BRANCH OF ALL-UNION RESEARCH INSTITUTE OF FERTILIZER, AGROTECHNICS AND AGRONOMICAL SOIL SCIENCE

| N                 | Animal ear tag number | Treated animals    |      |       |       |       |      |       |       |       |       |      |
|-------------------|-----------------------|--------------------|------|-------|-------|-------|------|-------|-------|-------|-------|------|
|                   |                       | Date of experiment |      |       |       |       |      |       |       |       |       |      |
|                   |                       | 29.05              | 5.06 | 13.06 | 26.06 | 27.06 | 4.07 | 24.07 | 30.07 | 20.08 | 29.08 | 1.10 |
| 1                 | 140                   | 0.29               | 0.24 | 0.37  | 0.21  | 0.21  | 0.14 | 0.36  | 0.01  | 0.10  | 0.17  | 0.03 |
| 2                 | 3169                  | 0.59               | 0.41 | 0.76  | 0.19  | 0.17  | 0.14 | 0.71  | 0.13  | 0.21  | 0.20  | 0.07 |
| 3                 | 9894                  | 0.32               | 0.26 | 0.37  | 0.34  | 0.12  | 0.16 | 0.23  | 0.14  | 0.08  | 0.06  | 0.20 |
| 4                 | 0703                  | 0.66               | 0.53 | 0.84  | 0.10  | 0.25  | 0.20 | -     | -     | -     | -     | -    |
| 5                 | 3182                  | 0.69               | 0.53 | 0.42  | 0.32  | 0.24  | 0.27 | 0.48  | 0.09  | 0.01  | 0.31  | 0.09 |
| 6                 | 0701                  | 1.02               | 0.99 | 1.17  | 0.21  | 0.23  | 0.37 | -     | -     | -     | -     | -    |
| 7                 | 0705                  | 0.69               | 0.49 | 0.38  | 0.20  | 0.20  | 0.16 | 0.58  | 0.09  | 0.01  | 0.18  | 0.29 |
| 8                 | 178                   | 0.17               | 0.24 | 0.83  | 0.14  | 0.52  | 0.38 | 0.93  | 0.05  | 0.01  | -     | -    |
| 9                 | 0720                  | 0.89               | 0.82 | 0.74  | 0.40  | 0.27  | 0.38 | -     | -     | -     | -     | -    |
| 10                | 166                   | 0.66               | 0.48 | 0.66  | 0.24  | 0.27  | -    | -     | -     | -     | -     | -    |
| Untreated animals |                       |                    |      |       |       |       |      |       |       |       |       |      |
| 16                | 3197                  | 2.62               | 1.78 | 1.24  | 0.93  | 0.65  | 0.40 | 1.26  | 0.44  | 0.24  | 0.80  | 0.41 |
| 17                | 67                    | 2.77               | 1.80 | 1.86  | 0.48  | 0.29  | 0.24 | 0.27  | 0.47  | 0.21  | 1.07  | 0.19 |
| 18                | 57                    | 2.87               | 1.82 | 1.91  | 0.86  | 0.39  | 0.46 | 1.18  | 0.14  | 0.12  | 0.93  | 0.34 |
| 19                | 249                   | 2.64               | 2.15 | 2.28  | 0.92  | 0.40  | 0.40 | 0.75  | 0.53  | 0.10  | 0.93  | 0.16 |
| 20                | 56                    | 2.79               | 2.24 | 2.46  | 0.65  | 0.53  | 0.47 | 0.97  | 0.33  | 0.27  | 0.70  | 0.25 |
| 21                | 121                   | 1.54               | 1.27 | 1.52  | 0.91  | 0.29  | 0.54 | 0.81  | 0.33  | 0.31  | 0.85  | 0.23 |
| 22                | 3121                  | 1.85               | 1.59 | 1.59  | 0.71  | 0.46  | 0.55 | 1.23  | 0.33  | 0.26  | 0.61  | 0.16 |
| 23                | 3141                  | 1.90               | 1.15 | 1.69  | 0.50  | 0.35  | 0.31 | 0.81  | 0.20  | 0.18  | 0.69  | -    |

TABLE B.5. MEAN DAILY MILK YIELD PER COW, L/d ( $\pm$  SD)  
IN PB TREATED AND UNTREATED COWS

| Date  | Treated        |       | Untreated       |     |
|-------|----------------|-------|-----------------|-----|
|       | L/d            | %     | L/d             | %   |
| 11-05 | 9.1 $\pm$ 0.9  | 100.0 | 10.0 $\pm$ 1.3  | 100 |
| 15-05 | 8.7 $\pm$ 0.8  | 95.6  | 10.01 $\pm$ 1.2 | 101 |
| 22-05 | 10.1 $\pm$ 1.1 | 110.9 | 11.0 $\pm$ 0.9  | 110 |
| 29-05 | 9.7 $\pm$ 0.6  | 106.6 | 10.3 $\pm$ 1.0  | 103 |
| 05-06 | 7.8 $\pm$ 0.7  | 85.7  | 8.6 $\pm$ 0.8   | 86  |
| 12-06 | 9.1 $\pm$ 0.5  | 100.0 | 10.7 $\pm$ 1.1  | 107 |
| 20-06 | 9.7 $\pm$ 1.0  | 106.0 | 9.4 $\pm$ 0.9   | 94  |
| 26-06 | 8.5 $\pm$ 0.9  | 93.4  | 9.9 $\pm$ 1.0   | 99  |
| 03-07 | 7.3 $\pm$ 0.5  | 80.2  | 9.8 $\pm$ 0.8   | 98  |
| 19-07 | 7.9 $\pm$ 0.6  | 86.8  | 8.5 $\pm$ 0.9   | 85  |
| 29-07 | 8.2 $\pm$ 0.8  | 90.1  | 9.1 $\pm$ 0.6   | 91  |
| 20-08 | 8.0 $\pm$ 0.6  | 87.9  | 8.5 $\pm$ 0.7   | 85  |
| 29-08 | 7.2 $\pm$ 0.5  | 79.1  | 7.0 $\pm$ 0.6   | 70  |
| 20-09 | 6.8 $\pm$ 0.6  | 74.7  | 7.3 $\pm$ 0.5   | 73  |

TABLE B.6. BLOOD PARAMETERS OF PB TREATED ANIMALS (MEAN  $\pm$  1 SD)

| Dates of tests                               | Erythrocytes<br>$\times 10^{12}/L$ | Leukocytes<br>$\times 10^9/L$ | Lymphocytes<br>(% leukocytes) | Basophils<br>(% leukocytes) | Monocytes<br>(% leukocytes) | Eosinophils<br>(% leukocytes) | Neutrophils<br>(% of leukocytes) |                |               |
|--|------------------------------------|-------------------------------|-------------------------------|-----------------------------|-----------------------------|-------------------------------|----------------------------------|----------------|---------------|
|  |                                    |                               |                               |                             |                             |                               | Mature                           | Band           | Young         |
| Prior to bolus administration                | 5.78 $\pm$ 0.3                     | 8.78 $\pm$ 0.71               | 52.0 $\pm$ 1.97               | 0                           | 4.4 $\pm$ 0.66              | 7.1 $\pm$ 0.98                | 35.7 $\pm$ 2.29                  | 1.0 $\pm$ 0.22 | 0             |
| One month later                              | 5.3 $\pm$ 0.4                      | 7.9 $\pm$ 0.6                 | 49.0                          | 0.4                         | 5.1 $\pm$ 0.7               | 6.7 $\pm$ 0.8                 | 35.3 $\pm$ 3.2                   | 2.3 $\pm$ 0.3  | 1.2 $\pm$ 0.4 |
| Two months after second bolus administration | 5.9 $\pm$ 0.4                      | 8.2 $\pm$ 0.7                 | 53.0 $\pm$ 2.1                | 0                           | 4.3 $\pm$ 0.48              | 5.4 $\pm$ 0.9                 | 34.3 $\pm$ 2.8                   | 2.4 $\pm$ 0.2  | 0.6 $\pm$ 0.4 |

TABLE B.7. THYROID HORMONE IN BLOOD SERUM, nmole/L

| Group     | T <sub>4</sub> | T <sub>3</sub> |
|-----------|----------------|----------------|
| Untreated | 47.2±5.3       | 1.8±0.16       |
| Treated   | 48.4±6.4       | 1.7±0.14       |

TABLE B.8. THIOCYANATE IN BLOOD PLASMA, nmole/L

| Group     | Thiocyanate content |
|-----------|---------------------|
| Untreated | 4.3±0.8             |
| Treated   | 4.1±0.9             |



TABLE B.9. TRACE- AND MACRO-ELEMENTS IN BLOOD SERUM OF CATTLE ( $\mu\text{g}/\text{Ml}$ )

| Group     | Micro- and macro-elements (mean $\pm$ S.D.) |                      |                     |                     |                      |                    |                      |                      |                        |                        |                        |      |
|-----------|---|----------------------|---------------------|---------------------|----------------------|--------------------|----------------------|----------------------|------------------------|------------------------|------------------------|------|
|           | K   | Na                   | Ca                  | Mg                  | Cu                   | Zn                 | Fe                   | Co                   | Zi                     | Mn                     | Cd                     | Pb   |
| Treated   | 205.0<br>$\pm 5.2$                          | 2945.0<br>$\pm 48.7$ | 49.7<br>$\pm 3.3$   | 17.96<br>$\pm 0.37$ | 0.785<br>$\pm 0.066$ | 0.74<br>$\pm 0.03$ | 1.944<br>$\pm 0.154$ | 0.024                | 0.0147<br>$\pm 0.0130$ | 0.0055<br>$\pm 0.0015$ | 0.0063<br>$\pm 0.0016$ | 0.04 |
| Untreated | 220.43<br>$\pm 7.98$                        | 2625.5<br>$\pm 99.0$ | 57.05<br>$\pm 2.77$ | 15.44<br>$\pm 0.48$ | 0.849<br>$\pm 0.070$ | 0.80<br>$\pm 0.06$ | 2.101<br>$\pm 0.515$ | 0.028<br>$\pm 0.008$ | 0.0033<br>$\pm 0.0007$ | 0.0092<br>$\pm 0.0027$ | 0.0131<br>$\pm 0.0011$ | 0.02 |

TABLE B.10. TRACE- AND MACRO-ELEMENT CONTENT OF BLOOD SERUM

| Cow<br>Ear Tag<br>No.   | Element concentration in serum $\mu\text{g}/\text{ml}$ |                       |                   |                     |                      |                    |                      |                      |                        |                        |                        |      |
|-------------------------|--|-----------------------|-------------------|---------------------|----------------------|--------------------|----------------------|----------------------|------------------------|------------------------|------------------------|------|
|                         | K  | Na                    | Ca                | Mg                  | Cu                   | Zn                 | Fe                   | Co                   | Li                     | Mn                     | Cd                     | Pb   |
| Treated                 |  |                       |                   |                     |                      |                    |                      |                      |                        |                        |                        |      |
| 140                     | 204.6  | 2 870.0               | 57.3              | 19.91               | 0.820                | 0.69               | 2.034                | -                    | no                     | 0.002                  | no                     | no   |
| 3169                    | 215.2  | 2 950.0               | 67.5              | 19.43               | 1.068                | 0.59               | 2.232                | -                    | -                      | 0.008                  | no                     | no   |
| 9894                    | 227.5  | 3 115.0               | 46.3              | 16.50               | 0.872                | 0.77               | 1.168                | no                   | 0.04                   | no                     | 0.008                  | no   |
| 0703                    | 190.5  | 3 135.0               | 58.2              | 17.99               | 0.616                | 0.75               | 1.894                | -                    | -                      | no                     | no                     | no   |
| 3182                    | 195.8  | 2 710.0               | 61.2              | 18.09               | 0.968                | 0.75               | 1.460                | no                   | -                      | no                     | 0.004                  | no   |
| 0701                    | 205.2  | 3 010.0               | 47.0              | 19.07               | 0.648                | 0.64               | 1.680                | no                   | -                      | no                     | 0.012                  | no   |
| 0705                    | 199.8  | 3 040.0               | 37.4              | 16.69               | 0.926                | -                  | 1.986                | no                   | -                      | 0.006                  | no                     | no   |
| 178                     | 227.0  | 2 690.0               | 42.8              | 17.64               | 0.888                | 0.88               | 2.520                | -                    | -                      | 0.006                  | 0.012                  | no   |
| 0720                    | 180.4  | 2 980.0               | 58.6              | 17.11               | 0.584                | 0.78               | 2.524                | -                    | 0.002                  | no                     | 0.002                  | no   |
| 166                     | 204.6  | 2 950.0               | 63.7              | 17.19               | 0.462                | 0.79               | -                    | 0.024                | 0.002                  | 0.004                  | 0.012                  | 0.04 |
| $\bar{x} \pm \text{SD}$ | 205.0<br>$\pm 5.2$                                     | 2 945.0<br>$\pm 48.7$ | 49.7<br>$\pm 3.3$ | 17.96<br>$\pm 0.37$ | 0.785<br>$\pm 0.066$ | 0.74<br>$\pm 0.03$ | 1.944<br>$\pm 0.154$ | 0.024                | 0.0147<br>$\pm 0.0130$ | 0.0055<br>$\pm 0.0015$ | 0.0063<br>$\pm 0.0016$ | 0.04 |
| Untreated               |  |                       |                   |                     |                      |                    |                      |                      |                        |                        |                        |      |
| 3197                    | 217.8  | 2 525.0               | 65.8              | 15.42               | 0.698                | 1.11               | 2.690                | -                    | -                      | 0.014                  | no                     | no   |
| 67                      | 220.8  | 3 020.0               | 46.7              | 16.10               | 0.980                | 0.95               | 5.644                | 0.048                | 0.004                  | 0.012                  | 0.018                  | no   |
| 57                      | 236.1  | 2 770.0               | 71.2              | 14.06               | 0.594                | 0.95               | 1.770                | 0.012                | 0.002                  | no                     | 0.012                  | no   |
| 249                     | 242.3  | 2 115.0               | 51.7              | 15.42               | 0.918                | 0.71               | 1.788                | -                    | -                      | no                     | no                     | no   |
| 56                      | 221.7  | 2 710.0               | 54.2              | 15.70               | 1.232                | 0.59               | 1.184                | -                    | -                      | 0.016                  | 0.010                  | no   |
| 121                     | 208.2  | 2 820.0               | 65.3              | 17.49               | 0.886                | 0.70               | 1.484                | 0.012                | -                      | no                     | 0.010                  | no   |
| 3121                    | 230.5  | 2 640.0               | 51.9              | 16.92               | 0.836                | 0.84               | 1.300                | 0.040                | 0.004                  | no                     | 0.012                  | 0.02 |
| 3141                    | 169.3  | 2 390.0               | 47.5              | 13.03               | 0.630                | 0.62               | 0.938                | no                   | no                     | 0.002                  | 0.014                  | no   |
| 183                     | 237.6  | 2 690.0               | 70.3              | 16.58               | 1.010                | 0.76               | 2.224                | -                    | -                      | no                     | 0.016                  | no   |
| 3174                    | 220.0  | 2 555.0               | 45.9              | 13.72               | 0.710                | 0.75               | 1.988                | -                    | no                     | 0.002                  | no                     | 0.02 |
| $\bar{x} \pm \text{SD}$ | 220.43<br>$\pm 8.00$                                   | 2 625.5<br>$\pm 99.0$ | 57.1<br>$\pm 2.8$ | 15.44<br>$\pm 0.48$ | 0.849<br>$\pm 0.070$ | 0.80<br>$\pm 0.06$ | 2.101<br>$\pm 0.515$ | 0.028<br>$\pm 0.008$ | 0.0033<br>$\pm 0.0007$ | 0.0092<br>$\pm 0.0027$ | 0.0131<br>$\pm 0.0011$ | 0.02 |

**THE EFFECTS OF MANURE FROM ANIMALS TREATED WITH  
FERROCYN-CONTAINING BOLI ON THE SOIL-PLANT  
TRANSFER OF RADIOCAESIUM (RUSSIAN STUDIES)**

**Lupin study**

The experiments with a leguminous fodder crop - European yellow lupin - were conducted under field conditions in 1991. The dung from cattle given ferrocyn-containing boli was used as an organic fertilizer. The potassium form of ferro-hexacyanoferrate (zeeom) was used in these studies:  $K_2Fe(CN)_6$ . The studies were carried out on soddy podzolic sandy soil (Table B.11.) on plots of 1 m<sup>2</sup> with 4 replicates of each of several treatments:

1. Control - no fertilizer applied
2. P<sub>60</sub>K<sub>60</sub> - mineral fertilizer, F (double superphosphate and agricultural salt) applied at 60 kg ha<sup>-1</sup>
3. P<sub>60</sub>K<sub>60</sub> + manure N 1 - mineral fertilizer and manure at a rate of 40 tonnes ha<sup>-1</sup> from **treated<sup>a</sup>** animals
4. P<sub>60</sub>K<sub>60</sub> + manure N 2 - mineral fertilizer and manure at a rate of 40 tonnes ha<sup>-1</sup> from **untreated<sup>b</sup>** animals
5. P<sub>60</sub>K<sub>60</sub> + zeeom - mineral fertilizer and zeeom (ferrocyn-containing compound based on sawdust)
6. zeeom

<sup>a</sup> Cs-137 activity in manure from treated animals was 2800 Bq kg<sup>-1</sup>.

<sup>b</sup> Cs-137 activity in manure from untreated animals was 2900 Bq kg<sup>-1</sup>.

Lupin was harvested at the stage of pod-formation, dried and ground in a mill and the <sup>137</sup>Cs activity measured.

**TABLE B.11. SOIL PARAMETERS ON WHICH EXPERIMENTS ON LUPIN WERE PERFORMED**

| Soil/parameters | pH  | P <sub>2</sub> O <sub>5</sub><br>(mg/100 g) | K <sub>2</sub> O<br>(mg/100 g) | Humus<br>(%) | Physical clay<br>(%) |
|-----------------|-----|---|--------------------------------|--------------|----------------------|
| Sand-podzolic   | 6.5 | 34.6  | 5.4                            | 1.9          | 4.7                  |

The results showed that <sup>137</sup>Cs content in green mass of lupin decreased by a factor of 1.6 compared with control treatment with mineral fertilizer applications. See Table B.12.

The positive effect of manure application (from ferrocyn treated cows) was noticed in the second year of the experiment (1992) with cocksfoot grasses (*Dactylis glomerata*) and timothy (*Phleum pratense*). Mineral fertilizers at a rate of N<sub>90</sub>P<sub>60</sub>K<sub>60</sub> were applied in treatments 2, 3 and 4 before sowing of grasses. Application of fertilizers at a rate of N<sub>90</sub>P<sub>60</sub>K<sub>60</sub> on soddy podzolic sandy soil reduced <sup>137</sup>Cs accumulation in grasses, in comparison with the treatment without fertilizers, by a factor of 1.6-1.9. Supplementary addition of manure (treatments 3 and 4) resulted in further significant reductions in <sup>137</sup>Cs accumulation in grasses in both first (3 and 1.5-fold respectively) and second (2 and 1-fold respectively) hay harvests.

Experiments involving different manure applications also illustrated the benefit of ferrocyn in both lupin and hay (Table B.13.).

TABLE B.12. SOIL-TO-PLANT AND SOIL-TO-GRASS (HAY) TRANSFER FACTORS [Bq/kg plant/kBq/m<sup>2</sup> soil] OF VARIOUS EXPERIMENTAL TREATMENTS ON ARABLE LAND WITH MANURE APPLICATIONS FROM DAIRY COWS GIVEN 3 BOLI

| Experimental treatment             | No fertilizers | F     | F + manure from control animals | F + manure of animals given AF CF | Statistical significance |
|------------------------------------|----------------|-------|---------------------------------|-----------------------------------|--------------------------|
|                                    | (1)            | (2)   | (4)                             | (3)                               |                          |
| 1991 Soil-lupin                    | 0.332          | 0.223 | 0.218                           | 0.137                             | 0.05                     |
| 1992 Soil-grass first hay harvest  | 0.404          | 0.216 | 0.150                           | 0.03                              | 0.03                     |
| 1992 Soil-grass second hay harvest | 0.559          | 0.346 | 0.225                           | 0.114                             | 0.07                     |

TABLE B.13. Cs-137 CONTENT IN SOIL AND GREEN MASS OF LUPIN

| Experimental treatment                     | Cs-137 content, Bq/kg |          | Transfer coefficient<br>[Bq/kg plant/kBq/m <sup>2</sup> soil] |
|--|-----------------------|----------|---|
|  | in soil               | in lupin |   |
| Control                                    | 1 220                 | 405      | 1.280   |
| P <sub>60</sub> K <sub>60</sub>            | 1 219                 | 272      | 0.806   |
| P <sub>60</sub> K <sub>60</sub> + manure 1 | 834                   | 182      | 0.840   |
| P <sub>60</sub> K <sub>60</sub> + manure 2 | 920                   | 126      | 0.530   |

Manure 1 - manure without ferrocyn, 40 t/ha

Manure 2 - manure with ferrocyn, 40 t/ha

It should be stressed that manure from animals given ferrocyn-containing boli was more effective in decreasing the transfer of <sup>137</sup>Cs to plants than manure from those not given boli. While the <sup>137</sup>Cs activity in lupin decreased 1.5 fold after the application of manure from untreated animals, manure from ferrocyn treated animals resulted in a 2.4 fold decrease.

Thus, manure from treated animals proved to be 62 % more effective in decreasing the <sup>137</sup>Cs transfer to plants compared with treatment involving the application of mineral (P and K) fertilizers only.

Evidently, ferrocyn-containing compounds administered to animals bind <sup>137</sup>Cs strongly and inhibit its transfer from manure to plants. Applying the artificial sorbent zeeom to soil decreased the <sup>137</sup>Cs transfer to lupin green mass 2.9 fold compared with the control.

Application of a complex of known agrochemical countermeasures together with the administration of sorbents of <sup>137</sup>Cs to livestock guarantees the production of clean milk, meat and fodder on territories with <sup>137</sup>Cs contamination up to 40 Ci/km<sup>2</sup>. <sup>137</sup>Cs accumulation in crops can be further reduced by applying manure from Prussian Blue administrated animals.

TABLE B.14. Cs-137 CONTENT IN SOIL AND COCKSFOOT (*Dactylis glomerata*) AND TIMOTHY (*Phleum pratense*), THE FIRST HAY HARVEST, 1992

| Experimental treatment                     | Cs-137 content, Bq/kg |            | Transfer coefficient<br>[Bq/kg plant/kBq/m <sup>2</sup> soil] |
|--|-----------------------|------------|---|
|  | in soil               | in grasses |   |
| Control                                    | 1 035                 | 419        | 1.35  |
| P <sub>60</sub> K <sub>60</sub>            | 1 150                 | 248        | 0.72  |
| P <sub>60</sub> K <sub>60</sub> + manure 1 | 845                   | 127        | 0.50  |
| P <sub>60</sub> K <sub>60</sub> + manure 2 | 1 089                 | 99         | 0.30  |

Manure 1 - manure without ferrocyn, 40 t/ha

Manure 2 - manure with ferrocyn, 40 t/ha

### Study with oats

With the objective of studying the effects of artificial and natural sorbents on <sup>137</sup>Cs transfer from soil to plants, investigations were performed under field conditions on the same soils as those with manure and ferrocyn-containing compounds described above. The sorbents tested were biefege, zeeom and vermiculite. Vermiculite, a natural sorbent, is a mica-like mineral formed from fieldspars and micas. Its chemical composition is unstable. The ferrocyn content in biefege was 10% and in zeeom 3%; in the trial, these two were equalized with respect to the active compound. The rate of application for biefege and vermiculite was 10 tons/ha; for zeeom it was 30 tons/ha. The experiment was designed with 4 replicates of each treatment on sites of 9 m<sup>2</sup>. The experimental plant was oats (Moscovsky-2 variety). The plants were grown to complete ripeness. After harvesting, the grain was ground by electrical grinder and counted for <sup>137</sup>Cs activity.

The experimental treatments were:

1. N<sub>60</sub>P<sub>60</sub>K<sub>60</sub>
2. N<sub>60</sub>P<sub>60</sub>K<sub>60</sub> + biefege
3. N<sub>60</sub>P<sub>60</sub>K<sub>60</sub> + zeeom
4. N<sub>60</sub>P<sub>60</sub>K<sub>60</sub> + vermiculite.

The experimental results showed that the various sorbents decreased <sup>137</sup>Cs transfer to oat grain 1.3-2.5 times. It should be noted that zeeom proved the most effective in decreasing <sup>137</sup>Cs transfer - see Table B.15.

TABLE B.15. THE EFFECT OF SORBENTS ON Cs-137 ACTIVITY IN OAT GRAIN

| Variants   | Transfer coefficient<br>[Bq/kg plant/kBq/m <sup>2</sup> soil] |
|--|---|
| N <sub>60</sub> P <sub>60</sub> K <sub>60</sub>                        | 0.20  |
| N <sub>60</sub> P <sub>60</sub> K <sub>60</sub> + biefege <sup>a</sup> | 0.12  |
| N <sub>60</sub> P <sub>60</sub> K <sub>60</sub> + zeeom                | 0.08  |
| N <sub>60</sub> P <sub>60</sub> K <sub>60</sub> + vermiculite          | 0.15  |
|  | SEM ± 0.12  |

<sup>a</sup> Biefege = Potassium ferric cyanoferrate (K Fe[Fe(CN)<sub>6</sub>])

## RESULTS OF STUDIES CONDUCTED BETWEEN 1993 AND 1995

In 1993 trials began on 4 collective farms in the south-western districts of the Bryansk region:

- i. "Majesky" - Klintsky district
- ii. "Novy Mir" - Klintsky district
- iii. "Smialtchsky" - Gordeevsky district
- iv. "Petrovo-bydsky" - Gordeevsky district.

The collective farm "Majesky" specialized in meat and milk production. The investigations were carried out on 180 Shvetskaya (Brown Swiss) cows situated on Veprin farm.

The collective farm "Novy Mir" specialized in meat and milk. The investigations were carried out on Unetcha farm on 201 Chernaya-pastraya (black and white) cows. Boli were given to 40 cows, ferrocyn powder was given to 81 cows, biefege was given to 40 cows and a control group of 40 cows was not given any treatment.

The collective farm "Smialtchsky" specialized in meat and milk. Experiments were carried out on 175 cows including a control group of 40 cows.

The collective farm "Petrovo-bydsky" also specialized in meat and milk production. Tests took place on the central farm with 100 cows.

Adult live weight of the cattle involved in the studies ranged from 350 to 500 kg. Milk production per cow ranged from 7-16 L/d. <sup>137</sup>Cs contamination levels of the farmland ranged between 15-40 Ci/km.

The trials were aimed at confirming the beneficial effects of PB 'on-farm'. Tables B.16. and B.17. illustrate the variable radiocaesium levels in pasture between farm locations and with month of year. Table B.18. shows the 3-fold reduction of milk radiocaesium levels within one week of providing PB.

TABLE B.16. Cs-137 CONTENT OF DIET OF COWS ON MAJESKY COLLECTIVE FARM, 1993

| Feed                                | Quantity in diet<br>kg | Cs-137 content, Bq/kg |         |
|-------------------------------------|------------------------|-----------------------|---------|
|                                     |                        | in 1 kg of fodder     | in diet |
| Green maize-silage                  | 20                     | 111                   | 2 220   |
| Rye-straw                           | 4                      | 222                   | 888     |
| Concentrates (rye,<br>oats, barley) | 3                      | 74                    | 222     |
| Brewers grains<br>(barda)           | 20                     | 7.4                   | 148     |
| Total:                              | 47                     |                       | 3 478   |

<sup>137</sup>Cs content in milk at the beginning of the experiment was  $358 \pm 25.9$  Bq/L

TABLE B.17. Cs-137 CONTENT (Bq/kg) IN FEED OFFERED TO DAIRY CATTLE ON COLLECTIVE FARM "NOVY MIR"

| Feed          | Sampling frequency (1994) |                  |                |                  |                |
|---------------|---------------------------|------------------|----------------|------------------|----------------|
|               | 03.08                     | 08.08            | 12.08          | 16.08            | 26.08          |
| Concentrates  | 111.0                     | 96.2             | 99.9           | 96.2             | 111.0          |
| Pasture grass | 2 625 ±<br>851            | 3 737 ±<br>1 184 | 3 293 ±<br>888 | 5 402 ±<br>1 221 | 5698 ±<br>1554 |

TABLE B.18. Cs-137 IN MILK, Bq/kg, FROM 'NOVY MIR' COLLECTIVE FARM

| Experimental treatment | Sampling dates (1994) |       |       |       |       |
|------------------------|-----------------------|-------|-------|-------|-------|
|                        | 03.08 <sup>a</sup>    | 08.08 | 12.08 | 16.08 | 26.08 |
| Control                | 185                   | 592   | 962   | 1 591 | 1 480 |
| Treated                | 222                   | 384   | 384   | 518   | 518   |

<sup>a</sup> Ferrocyn was administered to cows at a dose of 3-5 g/head/daily

TABLE B.19. Cs-137 CONTAMINATION LEVEL OF FEED AND MILK IN THE COLLECTIVE FARM 'SMIALTSCHKY' IN 1993

| Cs-137 content        | May-June      | July-August | September   |
|-----------------------|---------------|-------------|-------------|
| Pasture grass (Bq/kg) | 1 054.5 ± 157 | 765.9 ± 240 | 388.5 ± 148 |
| Diet (kBq/kg)         | 47.45         | 33.67       | 16.65       |
| Milk (Bq/L)           | 703 - 814     | 629 - 740   | 185 - 259   |

TABLE B.20. Cs-137 CONTENT IN FODDER AND MILK ON COLLECTIVE FARM 'PETROVO-BYDSKY' IN 1993

| Cs-137 content        | July | August | September |
|-----------------------|------|--------|-----------|
| Pasture grass (Bq/kg) | 888  | 1 406  | 1 036     |
| Milk (Bq/L)           | 407  | 518    | 444       |

#### The use of Prussian Blue in Russia

In 1994 Prussian Blue was licensed for use in Russia. In Table B.21. the total number of animals treated with PB is shown; these are divided into private farms and collective farms. Different ways of administrating PB are illustrated.

TABLE B.21. NUMBER OF ANIMALS GIVEN PRUSSIAN BLUE IN THE BRYANSK REGION IN 1994

| Type of administration          | Collective farm cattle | Private farm cattle |
|---------------------------------|------------------------|---------------------|
| Boli (ferrocyne)                | 1 717                  | 253                 |
| Bifege (10% PB + 90% cellulose) | 5 849                  | 1 244               |
| PB-powder                       | 4 240                  | 917                 |
| Salt licks                      | <u>406</u>             | <u>1 033</u>        |
| Total                           | 12 212                 | 3 442               |

TABLE B.22. REQUIREMENTS FOR PB COMPOUNDS IN RUSSIA IN 1995

| Forms           | Demand     | Total price<br>(million roubles) |
|-----------------|------------|----------------------------------|
| Bifege          | 15 tons    | 277                              |
| Boli            | 6 000 tons | 36                               |
| Ferrocyn powder | 2.3 tons   | 230                              |
| Salt licks      | 3.0 tons   | 15                               |

It was necessary to treat about 10 000 cows, 7000 on collective farms and 3000 on private farms.



## STUDIES PERFORMED IN THE UKRAINE

Experimental studies were carried out by the Ukrainian Institute of Agricultural Radiology (UIAR) at its experimental farm in the village of Polesskoe, Kiev region, on state and private farms in the village of Vilyun and on private farms in the villages of Lugovoe, Dubrovi district and Velikie Ozera, Rovno region.

Field studies were carried out on private farms in 3 districts of the Rovno Region in 1993 and 1 district of the Zhitomir region and 4 districts in the Rovno region in 1994.

### EXPERIMENTAL STUDIES

#### Materials and methods

On the experimental farms of UIAR two groups of 100 cows each were formed on 7 May 1991. Eighty-five test animals each received three 200 g boli containing 20% ferrocyn. Fifteen cows were treated with 3 ferrocyn boli containing additional microelements. After 2 months a further 2 boli were administered to each cow. The second group of 100 animals acted as control. The study lasted 5 months. Samples of forage were taken twice a month in order to assess the radionuclide content of the ration.

Ten cows were selected from both groups each month for measurement of their radiocaesium body content by live monitoring using a "Canberra-10" portable spectrometer. Once a week milk production was assessed and samples of milk sent for radiometrical and toxico-hygienic investigations.

Each month samples of peripheral blood were taken from the animals to determine haematological composition and microelement balance. In blood serum and plasma a number of biochemical parameters were measured: total protein and immunoglobulins, coefficient of antioxidant activity, activity of aspartate aminotransferase and alanine aminotransferase, concentration of malonic dialdehyde, amylase, cholesterol and ceruloplasmin.

Over the same period in the village of Vilyun, 20 animals on state farms and 8 animals on private farms each received 3 boli. In the villages of Lugovoye and Velikiye Ozera on 5 July 1991, 8 and 5 animals each received 2 and 3 boli respectively.

### RESULTS OF STUDIES BEGUN IN 1991

Cows from the UIAR experimental farm normally grazed the natural water meadows of the River Uzh. The extent of contamination of this pasture was 555 kBq/m (15 Ci/km<sup>2</sup>). The concentration of <sup>134</sup>Cs and <sup>137</sup>Cs in the grass varied from 120 to 1 200 Bq/kg (37-740 Bq/kg) depending on the exact location, and essentially did not change during the whole period of the experiment. Table B.23. presents approximate rations of the test cows and calculation of the daily intake of radionuclides by the animals. It shows that the major sources of <sup>134</sup>Cs and <sup>137</sup>Cs intake were hay and pasture grass. Calculation of the consumption of pasture forage indicated that cows ate from 10-16 kg dry matter/day.

Because the water meadows of the River Uzh (the main grazing areas) were flooded in May, cattle were obliged to graze the forest edges and glades, where the levels of soil contamination and radiocaesium concentration in grass was higher than on the main pasture. This explains the increase of radiocaesium concentration in cows milk in May and its decrease after the animals moved back to the water meadows.

In May, the daily intake of radiocaesium from forage was up to 20 kBq; in the following months of the experiment it decreased to 5-7 kBq. The <sup>137</sup>Cs concentration in the milk of treated cows was 1.5-4.5 times lower than that of the control group; the two values were 22-109 Bq/kg and 85-

165 Bq/kg, respectively (Table B.24.). In the experimental group that received additional microelements, radiocaesium concentrations in milk varied from 36-115 Bq/kg.

Haematological examination of the blood showed that the amount of leukocytes and erythrocytes in the control and experimental groups corresponded to physiological norms. No differences were found between the groups (Table B.25.).

The peripheral blood of cows was analysed for microelements. The data in Table B.26. demonstrate that there were no differences in microelement content between the control and experimental groups. The introduction of boli to cows did not influence the content of the above-mentioned microelements.

The results of the *in vivo* monitoring of radiocaesium content in the experimental animals (Table B.27.) showed that ferrocyn boli decreased the amount of  $^{137}\text{Cs}$  in body muscle by 5-23% in the treated cows compared with the control. Thus, in the treated group the  $^{137}\text{Cs}$  concentration decreased to 465 Bq/kg; in the control group, the lowest value recorded was 593 Bq/kg.

Initial investigations of the biochemical status demonstrated that there were no observable differences between the untreated and the treated groups (Table B.28.) over the first 90 days of PB treatment.

Biochemical investigations, carried out 6 months after the beginning of the experiment, did not detect any significant changes in the activity of the amino- and transaminating enzymes in liver and blood serum, the content of cholesterol, total lipids, beta-lipoproteins and glucose in blood and glycogen in liver homogenates. No significant changes in the activity of succinated dehydrogenase in liver mitochondria were observed. Parameters characterizing the kidneys' functional condition, viz., volume of urine excreted, its density and the urea content of blood serum and urine, all remained within physiological norms and did not differ from those of the control group.

The separate investigations of the immunological condition of the animals (i.e. phagocyte number, % phagocytic cells, bactericidal ability of blood and complement) did not show any statistically significant changes in these characteristics.

The weights of internal organs (liver, kidneys, heart, spleen, adrenals) were not different between the untreated and treated groups.

In Vilyun village (Dubrovich district, Rovno region) the cattle were grazed on pastures with contamination densities of about 370 kBq/m<sup>2</sup> (10 Ci/km<sup>2</sup>). The  $^{137}\text{Cs}$  concentration in pasture grass was 140-260 Bq/kg. The resulting milk radiocaesium levels are presented in Table B.29. From these data it is obvious that the levels of contamination in untreated cows' milk were 70-350 Bq/kg;  $^{137}\text{Cs}$  concentration in the milk of treated cows, over a period of 112 days, was 2-6 times lower than in the control group. The greatest difference in  $^{137}\text{Cs}$  levels occurred 35 days after introducing boli, when the level of radiocaesium in the milk of the treated cows was 4-6 times lower, than in the untreated group.

The *in vivo* assessment of tissue  $^{137}\text{Cs}$  content in cows showed that, 2 months after the beginning of the experiment, the radionuclide concentration in treated animals was 1.5 times lower than in the untreated group.

On private farms in the village of Vilyun, the  $^{137}\text{Cs}$  concentration in milk from treated cows decreased two-fold compared with the untreated group (Table B.30.). The animals were grazed on pasture with a soil contamination of 248 kBq/m<sup>2</sup> (6.7 Ci/km<sup>2</sup>).

In Lugovoye village, cows from private farms grazed pastures with a contamination of 648 kBq/m<sup>2</sup> (17.5 Ci/km<sup>2</sup>).  $^{137}\text{Cs}$  levels in grass were 7.5-9.8 kBq/kg. Analyses showed (Table B.31.)

that the milk of experimental cows had a radiocaesium content 2 times lower than the control group. However, the introduction of two boli in this experiment did not decrease milk levels to below 370 Bq/L.

In the village of Velikie Ozero, cows from private farms grazed pastures with a radiocaesium level of about 130 kBq/m<sup>2</sup> (3.5 Ci/km<sup>2</sup>), which was considerably lower than the other villages in the experiment. However, a high level of contamination was found in the milk of the control group (900-1300 Bq/L); this was probably due to the very high coefficient of transfer in the soil-grass chain. The application of 3 boli in the experiment made it possible to reduce the milk <sup>137</sup>Cs concentration (Table B.32.), and to provide milk with a radiocaesium level lower than the TPL of 370 Bq/L.

TABLE B.23. CALCULATED DAILY INTAKE OF RADIOCAESIUM FROM GRASS/FORAGE CONSUMED BY COWS ON THE EXPERIMENTAL FARM OF UKRAINIAN RESEARCH INSTITUTE OF AGRICULTURAL RADIOLOGY

| Forage                                  | May                      |                        |                          | June - September         |                        |                          |
|---|--------------------------|------------------------|--------------------------|--------------------------|------------------------|--------------------------|
|   | Daily consumption<br>kg  | Radiocaesium<br>kBq/kg | Content in forage<br>kBq | Daily consumption,<br>kg | Radiocaesium<br>Kbq/kg | Content in forage<br>kBq |
| Pasture grass                           | 10 - 16                  | 0.57 - 0.74            | up to 11.84              | 10 - 16                  | 0.5 - 0.8              | to 12.8                  |
| Green mass of sown<br>grasses and maize | 20                       | 0.18                   | 3.6                      | 25                       | 0.01 - 0.18            | 0.25 - 4.5               |
| Hay of cereal and<br>leguminous crops   | 2                        | 2.22                   | 4.44                     | -                        | -                      | -                        |
| Mixed forage                            | 3                        | 0.004                  | 0.01                     | 3                        | 0.004                  | 0.01                     |
| Total in ration                         | 13.5 - 14.0 <sup>a</sup> | -                      | up to 20.00              | up to 14.0*              | -                      | up to 17.3               |

<sup>a</sup> dry matter

TABLE B.24. Cs-137 CONCENTRATION IN THE MILK OF COWS ON THE EXPERIMENTAL FARM OF THE UKRAINIAN RESEARCH INSTITUTE OF AGRICULTURAL RADIOLOGY IN THE VILLAGE OF POLESSKOYE (Bq/L), MEAN  $\pm$  SD

| Groups of animals         | Initial data | Days after the beginning of the experiment |              |               |               |               |               |              |              |              |               |               |               |               |               |               |               |               |               |
|---------------------------|--------------|--|--------------|---------------|---------------|---------------|---------------|--------------|--------------|--------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|
|                           |              | 7  | 14           | 21            | 35            | 42            | 49            | 63           | 70           | 77           | 84            | 91            | 105           | 112           | 119           | 126           | 133           | 140           | 147           |
| Control                   | 76.0         | 98.0                                       | 183.0        | 258.0         | 104.0         | 94.0          | 104.0         | 88.0         | 114.0        | 97.0         | 104.0         | 121.0         | 147.0         | 112.0         | 117.0         | 131.0         | 113.0         | 115.0         | 165.0         |
|                           | $\pm$<br>3.7 | $\pm$<br>5.1                               | $\pm$<br>8.8 | $\pm$<br>15.9 | $\pm$<br>11.8 | $\pm$<br>10.0 | $\pm$<br>10.4 | $\pm$<br>4.9 | $\pm$<br>9.9 | $\pm$<br>4.5 | $\pm$<br>18.5 | $\pm$<br>14.6 | $\pm$<br>15.7 | $\pm$<br>9.7  | $\pm$<br>14.1 | $\pm$<br>10.7 | $\pm$<br>10.1 | $\pm$<br>10.5 | $\pm$<br>20.4 |
| Experiment 1 <sup>a</sup> | 68.0         | 22.0                                       | 37.0         | 63.0          | 53.0          | 56.0          | 58.0          | 43.0         | 51.0         | 64.0         | 71.0          | 106.0         | 95.0          | 63.0          | 77.0          | 93.0          | 88.0          | 109.0         | 64.0          |
|                           | $\pm$<br>4.9 | $\pm$<br>1.8                               | $\pm$<br>2.0 | $\pm$<br>4.4  | $\pm$<br>7.4  | $\pm$<br>7.7  | $\pm$<br>6.8  | $\pm$<br>2.4 | $\pm$<br>5.2 | $\pm$<br>5.1 | $\pm$<br>3.7  | $\pm$<br>7.2  | $\pm$<br>6.5  | $\pm$<br>6.2  | $\pm$<br>6.5  | $\pm$<br>14.2 | $\pm$<br>6.7  | $\pm$<br>10.5 | $\pm$<br>7.4  |
| Experiment 2 <sup>b</sup> | 69.0         | -  | -            | -             | -             | 85.0          | -             | -            | 66.0         | -            | -             | 115.0         | -             | 65.0          | -             | 99.0          | -             | -             | 92.0          |
|                           | $\pm$<br>5.2 | -  | -            | -             | -             | $\pm$<br>5.8  | -             | -            | $\pm$<br>5.8 | -            | -             | $\pm$<br>13.1 | -             | $\pm$<br>10.0 | -             | $\pm$<br>8.1  | -             | -             | $\pm$<br>14.4 |

<sup>a</sup> 3 boli; 2 units were introduced again after 2 months

<sup>b</sup> boli with microelements

TABLE B.25. HAEMATOLOGICAL CHARACTERISTICS (MEAN  $\pm$  SD) OF TREATED AND UNTREATED COWS ON THE EXPERIMENTAL FARM OF THE UKRAINIAN RESEARCH INSTITUTE OF AGRICULTURAL RADIOLOGY

| Group of animals          | Days from application of boli |                               |                          |                               |                          |                               |                          |                               |                          |                               |
|---------------------------|-------------------------------|-------------------------------|--------------------------|-------------------------------|--------------------------|-------------------------------|--------------------------|-------------------------------|--------------------------|-------------------------------|
|                           | 0                             |                               | 30                       |                               | 60                       |                               | 90                       |                               | 120                      |                               |
|                           | Leuk.<br>$\times 10^9/L$      | Erythr.<br>$\times 10^{12}/L$ | Leuk.<br>$\times 10^9/L$ | Erythr.<br>$\times 10^{12}/L$ | Leuk.<br>$\times 10^9/L$ | Erythr.<br>$\times 10^{12}/L$ | Leuk.<br>$\times 10^9/L$ | Erythr.<br>$\times 10^{12}/L$ | Leuk.<br>$\times 10^9/L$ | Erythr.<br>$\times 10^{12}/L$ |
| Control                   | 7.76                          | 4.21                          | 9.74                     | 4.36                          | 8.04                     | 4.07                          | 10.25                    | 5.20                          | 9.92                     | 5.22                          |
|                           | $\pm$                         | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         |
|                           | 1.73                          | 0.33                          | 1.36                     | 0.07                          | 2.02                     | 0.17                          | 1.84                     | 0.19                          | 1.61                     | 0.15                          |
| Experiment 1 <sup>a</sup> | 6.12                          | 4.56                          | 6.96                     | 4.33                          | 7.00                     | 4.08                          | 7.12                     | 4.96                          | 6.67                     | 5.02                          |
|                           | $\pm$                         | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         |
|                           | 0.44                          | 0.11                          | 0.69                     | 0.12                          | 0.51                     | 0.08                          | 0.54                     | 0.19                          | 0.55                     | 0.23                          |
| Experiment 2 <sup>b</sup> | 6.12                          | 4.56                          | 8.32                     | 4.17                          | 8.04                     | 4.24                          | 8.28                     | 4.80                          | 8.20                     | 5.12                          |
|                           | $\pm$                         | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         | $\pm$                    | $\pm$                         |
|                           | 0.44                          | 0.11                          | 0.93                     | 0.08                          | 0.61                     | 0.09                          | 0.88                     | 0.27                          | 0.83                     | 0.30                          |

<sup>a</sup> "pure" boli

<sup>b</sup> boli with additional microelements

TABLE B.26. MICROELEMENT CONTENT OF BLOOD IN CONTROL AND EXPERIMENTAL COWS ON THE EXPERIMENTAL FARM OF THE UKRAINIAN RESEARCH INSTITUTE OF AGRICULTURAL RADIOLOGY (mg/100 ml), MEAN  $\pm$  SD

| Micro-element | Groups of animals |         |         |         |                           |         |         |         |                           |         |         |         |
|---------------|-------------------|---------|---------|---------|---------------------------|---------|---------|---------|---------------------------|---------|---------|---------|
|               | Control           |         |         |         | Experiment 1 <sup>a</sup> |         |         |         | Experiment 2 <sup>b</sup> |         |         |         |
|               | Initial value     | 1 month | 2 month | 3 month | Initial value             | 1 month | 2 month | 3 month | Initial value             | 1 month | 2 month | 3 month |
| Mn            | 12.48             | 17.38   | 13.95   | 12.60   | 12.48                     | 18.08   | 18.20   | 12.86   | 12.48                     | 16.14   | 28.82   | 15.74   |
|               | $\pm$             | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   |
|               | 1.10              | 1.65    | 4.35    | 3.37    | 1.10                      | 1.44    | 1.83    | 0.96    | 1.10                      | 1.32    | 8.54    | 0.75    |
| Fe            | 69.80             | 82.62   | 58.15   | 64.30   | 69.80                     | 85.78   | 78.46   | 64.24   | 69.80                     | 82.92   | 90.62   | 87.92   |
|               | $\pm$             | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   |
|               | 6.18              | 3.11    | 14.06   | 16.25   | 16.18                     | 9.91    | 9.44    | 6.23    | 6.18                      | 6.12    | 10.80   | 4.08    |
| Cu            | 0.54              | 0.28    | -       | 0.21    | 0.54                      | 0.47    | -       | 0.23    | 0.54                      | 0.23    | -       | 0.23    |
|               | $\pm$             | $\pm$   |         | $\pm$   | $\pm$                     | $\pm$   |         | $\pm$   | $\pm$                     | $\pm$   |         | $\pm$   |
|               | 0.15              | 0.05    | -       | 0.03    | 0.15                      | 0.14    | -       | 0.04    | 0.15                      | 0.04    | -       | 0.02    |
| Zn            | 0.44              | 0.3     | -       | 0.27    | 0.44                      | 0.53    | -       | 0.28    | 0.44                      | 0.31    | -       | 0.33    |
|               | $\pm$             | $\pm$   |         | $\pm$   | $\pm$                     | $\pm$   |         | $\pm$   | $\pm$                     | $\pm$   |         | $\pm$   |
|               | 0.08              | 0.06    | -       | 0.08    | 0.08                      | 0.09    | -       | 0.05    | 0.08                      | 0.07    | -       | 0.05    |
| Mo            | 0.02              | 0.05    | 0.03    | 0.04    | 0.02                      | 0.06    | 0.04    | 0.04    | 0.02                      | 0.05    | 0.06    | 0.07    |
|               | $\pm$             | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   |
|               | 0.002             | 0.01    | 0.009   | 0.01    | 0.002                     | 0.01    | 0.005   | 0.008   | 0.002                     | 0.01    | 0.02    | 0.01    |
| Co            | 0.05              | 0.07    | 0.04    | 0.05    | 0.05                      | 0.07    | 0.06    | 0.06    | 0.05                      | 0.07    | 0.09    | 0.07    |
|               | $\pm$             | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   | $\pm$                     | $\pm$   | $\pm$   | $\pm$   |
|               | 0.01              | 0.008   | 0.009   | 0.009   | 0.01                      | 0.007   | 0.02    | 0.009   | 0.01                      | 0.009   | 0.01    | 0.008   |

<sup>a</sup> "pure" boli (5 animals per group)

<sup>b</sup> boli with additional microelements

TABLE B.27. DETERMINATION OF THE RADIOCAESIUM CONTENT IN MUSCLE *in vivo* OF EXPERIMENTAL ANIMALS BY USE OF A "CANBERRA-10" SPECTROMETER ON THE EXPERIMENTAL FARM OF THE UKRAINIAN RESEARCH INSTITUTE OF AGRICULTURAL RADIOLOGY (Bq/kg), MEAN  $\pm$  SD

| Groups of animals <sup>a</sup> | Initial data | Days after the beginning of the experiment |       |       |       |       |       |       |       |
|--------------------------------|--------------|--|-------|-------|-------|-------|-------|-------|-------|
|                                |              | 10   | 24    | 39    | 53    | 67    | 100   | 130   | 147   |
| Untreated                      | 543.0        | 601.0                                      | 635.5 | 620.2 | 593.0 | 595.3 | 592.6 | 596.9 | 601.5 |
|                                | $\pm$        | $\pm$                                      | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ |
|                                | 48.9         | 41.3                                       | 41.8  | 43.7  | 31.3  | 42.3  | 51.7  | 55.9  | 63.6  |
| Treated                        | 562.0        | 508.2                                      | 487.2 | 475.2 | 495.1 | 464.7 | 511.1 | 567.0 | 581.4 |
|                                | $\pm$        | $\pm$                                      | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ |
|                                | 46.6         | 56.9                                       | 42.6  | 42.3  | 54.9  | 44.5  | 52.3  | 62.5  | 62.9  |

<sup>a</sup> 10 animals per group



TABLE B.28. BIOCHEMICAL CHARACTERISTICS (MEAN  $\pm$  SD) OF BLOOD PLASMA IN COWS ON THE EXPERIMENTAL FARM OF THE UKRAINIAN RESEARCH INSTITUTE OF AGRICULTURAL RADIOLOGY

| Animal groups             | Malonic dialdehyde<br>un.OD $\times$ 1000 | K anti-oxidative activity,<br>rel.un. | Ceruloplasmin<br>mg % | Immuno-globulins<br>g/L | Amylase<br>mg/ml | Cholesterol<br>mg % | ASAT<br>$\mu$ mol/ml/hr | ACAT<br>$\mu$ mol/ml/hr |
|---------------------------|---|---------------------------------------|-----------------------|-------------------------|------------------|---------------------|-------------------------|-------------------------|
| Before experiment         |   |                                       |                       |                         |                  |                     |                         |                         |
| Control                   | 27.0 $\pm$ 8.0                            | 1.16 $\pm$ 0.07                       | -                     | 17.9 $\pm$ 0.1          | 48.8 $\pm$ 3.5   | 198 $\pm$ 40.5      | 2.86 $\pm$ 0.19         | 0.97 $\pm$ 0.06         |
| Experiment                | 26.4 $\pm$ 3.6                            | 1.07 $\pm$ 0.07                       | -                     | 16.8 $\pm$ 0.3          | 38.9 $\pm$ 2.8   | 239 $\pm$ 13.0      | 2.77 $\pm$ 0.08         | 0.97 $\pm$ 0.06         |
| Day 30 of experiment      |   |                                       |                       |                         |                  |                     |                         |                         |
| Control                   | 28.4 $\pm$ 4.3                            | 1.17 $\pm$ 0.06                       | 7.4 $\pm$ 0.58        | 16.6 $\pm$ 0.3          | 22.9 $\pm$ 5.2   | 210 $\pm$ 30.8      | 2.04 $\pm$ 0.19         | 0.97 $\pm$ 0.06         |
| Experiment 1 <sup>a</sup> | 31.4 $\pm$ 2.7                            | 1.22 $\pm$ 0.08                       | 8.5 $\pm$ 0.88        | 17.0 $\pm$ 0.04         | 36.7 $\pm$ 3.6   | 210 $\pm$ 18.7      | 1.75 $\pm$ 0.15         | 0.52 $\pm$ 0.06         |
| Experiment 2 <sup>b</sup> | 27.0 $\pm$ 2.7                            | 1.12 $\pm$ 0.03                       | 7.8 $\pm$ 0.53        | 16.6 $\pm$ 0.6          | 21.8 $\pm$ 6.0   | 212 $\pm$ 25.3      | 2.00 $\pm$ 0.11         | 0.46 $\pm$ 0.10         |
| Day 60 of experiment      |   |                                       |                       |                         |                  |                     |                         |                         |
| Control                   | 25.8 $\pm$ 4.3                            | 1.62 $\pm$ 0.20                       | 7.3 $\pm$ 1.26        | 16.4 $\pm$ 0.8          | 25.2 $\pm$ 3.1   | 156 $\pm$ 21.2      | 2.01 $\pm$ 0.14         | 0.80 $\pm$ 0.10         |
| Experiment 1 <sup>a</sup> | 29.2 $\pm$ 1.9                            | 1.52 $\pm$ 0.06                       | 8.5 $\pm$ 1.20        | 16.9 $\pm$ 0.0          | 29.9 $\pm$ 6.5   | 206 $\pm$ 17.5      | 2.08 $\pm$ 0.17         | 0.72 $\pm$ 0.10         |
| Experiment 2 <sup>b</sup> | 33.6 $\pm$ 4.1                            | 1.59 $\pm$ 0.09                       | 7.6 $\pm$ 0.51        | 17.4 $\pm$ 0.8          | 31.7 $\pm$ 9.6   | 187 $\pm$ 21.1      | 2.28 $\pm$ 0.16         | 0.78 $\pm$ 0.16         |
| Day 90 of experiment      |   |                                       |                       |                         |                  |                     |                         |                         |
| Control                   | 25.8 $\pm$ 4.3                            | 1.58 $\pm$ 0.08                       | 5.6 $\pm$ 0.78        | 17.4 $\pm$ 0.0          | 49.1 $\pm$ 4.7   | 137 $\pm$ 24.2      | 1.95 $\pm$ 0.12         | 1.15 $\pm$ 0.09         |
| Experiment 1 <sup>a</sup> | 35.1 $\pm$ 1.8                            | 1.56 $\pm$ 0.07                       | 5.9 $\pm$ 0.53        | 17.6 $\pm$ 0.3          | 39.1 $\pm$ 2.7   | 173 $\pm$ 5.9       | 2.19 $\pm$ 0.10         | 0.71 $\pm$ 0.07         |
| Experiment 2 <sup>b</sup> | 36.6 $\pm$ 3.4                            | 1.42 $\pm$ 0.06                       | 6.8 $\pm$ 0.71        | 17.1 $\pm$ 0.4          | 56.2 $\pm$ 4.6   | 185 $\pm$ 19.9      | 1.99 $\pm$ 0.07         | 0.75 $\pm$ 0.15         |

<sup>a</sup> "pure" boli

<sup>b</sup> boli with microelements

TABLE B.29. Cs-137 CONCENTRATION IN THE MILK OF COWS ON COLLECTIVE FARM, "CHAPAYEV", IN THE VILLAGE OF VILYUN (Bq/L), MEAN  $\pm$  SD

| Groups of animals       | Initial data | Days after the beginning of the experiment |       |       |       |       |       |       |       |       |       |       |       |       |       |       |       |
|-------------------------|--------------|--|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
|                         |              | 7  | 14    | 21    | 28    | 35    | 42    | 48    | 56    | 63    | 70    | 77    | 84    | 91    | 98    | 105   | 112   |
| Control                 | 145.9        | 90.0                                       | 84.6  | 99.0  | 78.7  | 115.8 | 79.6  | 70.8  | 154.6 | 181.0 | 216.2 | 299.8 | 323.9 | 198.8 | 291.8 | 347.6 | 264.1 |
|                         | $\pm$        | $\pm$                                      | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ |
|                         | 15.6         | 6.0  | 6.7   | 10.3  | 6.1   | 10.3  | 7.1   | 4.5   | 10.6  | 10.2  | 14.3  | 15.4  | 17.6  | 10.8  | 13.7  | 13.4  | 22.7  |
| Experiment <sup>a</sup> | 105.5        | 22.9                                       | 14.8  | 18.5  | 13.5  | 24.3  | 33.3  | 60.1  | 56.1  | 41.2  | 59.7  | 61.1  | 93.9  | 81.8  | 183.5 | 184.2 | 158.3 |
|                         | $\pm$        | $\pm$                                      | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ |
|                         | 12.3         | 1.5  | 4.4   | 4.2   | 2.4   | 2.0   | 2.6   | 3.6   | 3.4   | 2.7   | 4.3   | 3.5   | 7.2   | 3.8   | 11.1  | 39.5  | 16.7  |

<sup>a</sup> 3 boli were introduced per os, on day 0 and 2 more boli at day 60

TABLE B.30. Cs-137 CONCENTRATION IN THE MILK OF COWS ON PRIVATE FARMS IN THE VILLAGE OF VILYUN (Bq/L), MEAN  $\pm$  SD

| Groups of animals       | Initial data | Days after the beginning of the experiment |       |       |       |       |       |       |       |       |       |       |       |
|-------------------------|--------------|--|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|-------|
|                         |              | 7  | 14    | 21    | 28    | 35    | 42    | 49    | 56    | 63    | 70    | 77    | 84    |
| Control                 | 58.3         | 23.7                                       | 33.8  | 23.5  | 18.0  | 24.9  | 21.9  | 30.7  | 19.9  | 19.7  | 19.6  | 52.2  | 54.4  |
|                         | $\pm$        | $\pm$                                      | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ |
|                         | 21.6         | 5.3  | 2.3   | 2.5   | 1.4   | 1.7   | 3.4   | 3.9   | 1.3   | 2.5   | 2.0   | 2.7   | 3.3   |
| Experiment <sup>a</sup> | 62.6         | 23.3                                       | 17.7  | 11.8  | 10.6  | 13.8  | 12.8  | 24.4  | 23.5  | 13.3  | 18.8  | 49.7  | 48.1  |
|                         | $\pm$        | $\pm$                                      | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ | $\pm$ |
|                         | 12.0         | 4.4  | 2.2   | 1.9   | 1.5   | 1.2   | 1.3   | 3.1   | 1.4   | 2.4   | 1.9   | 3.3   | 1.1   |

<sup>a</sup> 3 boli were given per os on day 0

TABLE B.31. Cs-137 CONCENTRATION IN THE MILK OF COWS ON PRIVATE FARMS IN THE VILLAGE OF LUGOVOYE (Bq/L), MEAN  $\pm$  SD

| Groups of animals       | Initial data  | Days after the beginning of the experiment |               |               |               |               |               |               |               |               |               |               |
|-------------------------|---------------|--|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|
|                         |               | 7  | 14            | 21            | 28            | 35            | 42            | 49            | 56            | 63            | 70            | 77            |
| Control                 | 887.2         | 845.1                                      | 1053.4        | 1048.1        | 941.3         | 678.7         | 940.3         | 920.4         | 763.4         | 881.8         | 582.8         | 616.7         |
|                         | $\pm$<br>41.3 | $\pm$<br>83.1                              | $\pm$<br>28.8 | $\pm$<br>41.2 | $\pm$<br>33.6 | $\pm$<br>41.4 | $\pm$<br>65.2 | $\pm$<br>37.8 | $\pm$<br>43.9 | $\pm$<br>72.5 | $\pm$<br>29.6 | $\pm$<br>35.2 |
| Experiment <sup>a</sup> | 1005.3        | 602.3                                      | 673.4         | 620.5         | 490.1         | 338.8         | 539.4         | 567.0         | -             | 586.4         | 368.6         | 379.3         |
|                         | $\pm$<br>63.3 | $\pm$<br>58.6                              | $\pm$<br>66.4 | $\pm$<br>56.6 | $\pm$<br>55.0 | $\pm$<br>47.8 | $\pm$<br>49.8 | $\pm$<br>62.1 | -             | $\pm$<br>55.2 | $\pm$<br>31.6 | $\pm$<br>38.2 |

<sup>a</sup> 2 boli were introduced per os on day 0

TABLE B.32. Cs-137 CONCENTRATION IN THE MILK OF COWS ON PRIVATE FARMS IN THE VILLAGE OF VELIKIYE OZERA (Bq/L), MEAN  $\pm$  SD

| Groups of animals       | Initial data  | Days after the beginning of the experiment |               |               |               |               |               |               |               |               |               |               |               |
|-------------------------|---------------|--|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|---------------|
|                         |               | 7  | 14            | 21            | 28            | 35            | 42            | 49            | 56            | 70            | 77            | 84            | 91            |
| Control                 | 1508.4        | 826.7                                      | 1086.4        | 1281.9        | 1207.8        | 1163.8        | 1192.1        | 1107.0        | 1150.0        | 1354.2        | 1169.2        | 1184.0        | 754.8         |
|                         | $\pm$<br>38.0 | $\pm$<br>56.3                              | $\pm$<br>82.1 | $\pm$<br>30.3 | $\pm$<br>58.4 | $\pm$<br>38.1 | $\pm$<br>44.4 | $\pm$<br>37.2 | $\pm$<br>50.6 | $\pm$<br>77.2 | $\pm$<br>89.2 | $\pm$<br>65.5 | $\pm$<br>38.1 |
| Experiment <sup>a</sup> | 1262.2        | 295.0                                      | 314.7         | 319.9         | 373.9         | 300.4         | 313.6         | 307.5         | 290.9         | 454.0         | 359.0         | 367.2         | 303.4         |
|                         | $\pm$<br>68.8 | $\pm$<br>43.0                              | $\pm$<br>24.0 | $\pm$<br>53.7 | $\pm$<br>63.3 | $\pm$<br>37.4 | $\pm$<br>37.4 | $\pm$<br>49.2 | $\pm$<br>22.0 | $\pm$<br>39.1 | $\pm$<br>40.9 | $\pm$<br>51.2 | $\pm$<br>38.5 |

<sup>a</sup> 3 boli were introduced per os on day 0

## RESULTS OF FIELD STUDIES CARRIED OUT IN 1993/1994

Field studies on the use of Prussian Blue were carried out on over 6600 cows kept by private farmers in 90 contaminated village settlements in the Rovno and Zhitomir regions during the grazing periods (late April - mid-October) in 1993 and 1994. A typical farmer has 1-2 cows who graze common pastures and forest clearings. Average yield of these small cattle ( $395 \pm 17$  kg body weight) is  $3400 \pm 120$  litres/lactation. Radiocaesium levels in typical pasture and milk samples from such farms at the beginning of the grazing season were 1150-1912 Bq/kg and 640 -960 Bq/L respectively. The levels of radiocaesium in soils is dependent on the type of soil viz. Zhitomir region (soddy podzolic soils) 74-555 Kbk/m<sup>2</sup>; Rovno region (peaty soils) 37-185 Kbk/m<sup>2</sup>.

The field study farms are located in the following villages:

1993

### ROVNO REGION

#### Dubrovitsa district

Velikiye Ozera  
Makhi  
Rizki  
Budimlya  
Chermen

#### Zarichnoye

Lisichin  
Ostritsi  
Tikhovin  
Parskoye  
Boriva

#### Rokitnoye district

Budki Kamyanystye  
Vizhitsa  
Zalavye  
Drozdil  
Elno  
Perekhodichi  
Staroye Selo  
Ostritsk  
Zabolotnoye  
Tomashgorod

### ZHITOMIR REGION

#### Ovruch district

Kamyanyivka  
Rudnya  
Pavluykivka  
Mogulnya  
Semeny

Byurin  
Sloboda  
Rokitno  
Stovnechno  
Mliny

Usovo  
Lymany  
Doneta  
Gorodets  
Chervonosilye

1994

### ZHITOMIR REGION

#### Ovruch district

Bigun  
Selezivka  
Krasilovka  
Listvin  
Vozlyakove  
Polokhachev  
Pribitki  
Kozuli

Sholomky  
Antonovichy  
Zadorozhnoyea  
Verkhnya Rudnya  
Koraki  
Sirkivschina  
Krasnosilka  
Pobichy

Duby  
Nizhnya Rudnya  
Olenichy  
Perebrody  
Ignatpil  
Mala Fosnya

### ROVNO REGION

#### Volodimirets district

Dubrovits district  
Zhaden  
Velikiye Ozera  
Veliky Chermen  
Milayachy

Beloye  
Luki  
Velikiye  
Telkovitchy  
Zhuravlino

Bererno  
Zirka  
Uricchya  
Velikiya Temkovichy

## ROVNO REGION

| Rokitnoye district |           | Zarichnoye district |
|--------------------|-----------|---------------------|
| Budki Kamyanystye  | Osnitsk   | Bir                 |
| Drozdin            | Zalovya   | Zelenaya            |
| Staroye Selo       | Bilsk     | Dibrova             |
| Velshitsa          | Elno      | Mlinok              |
| Perekhodichi       | Gribun    | Ostrizk             |
| Borova             | Zabolotya | Parska              |
| Masovychy          |           | Vichki              |
|                    |           | Tikhovisk           |
|                    |           | Osichin             |

During 1993 boli were administered to a total of 1 252 cows (3 per animal) and 220 animals received ferrocyn powder. In 1994 a total of 5 141 cows on farms in 70 villages received either boli or ferrocyn added to concentrate (4 401 cows and 740 animals received boli or ferrocyn treated forage, respectively).

### RESULTS

The results of the field studies can be exemplified by those obtained in the village of Velikiye Ozera, Dubrovitsa district, Rovno region. In this village, 130 cows received boli (3 per animal) and 75 animals served as controls. All grazed the same pasture. After a grazing period of 40 days the milk radiocaesium concentrations were:

|                    |                |
|--------------------|----------------|
| Control animals    | 534 ± 37 Bq/L  |
| Bolus-treated cows | 237 ± 25 Bq/L. |

Viz. a two-fold reduction in milk radiocaesium concentration.

Although the detailed data are not given for all animals, the overall effect of using PB boli was to reduce the milk radiocaesium concentration by a factor of 2-4. Adding ferrocyn powder to the diet had a similar effect; in this case the reduction achieved was 2.5 to 4.2 fold.

The effects of bolus application on radiocaesium content of meat was assessed in 20 animals; 3-6 fold reductions were recorded.

### CONCLUSIONS

- (1) The use of Prussian Blue (ferrocyn) as either boli or direct application to forage decreased the radiocaesium content of both milk and meat.
- (2) Application of 2 ferrocyn containing boli resulted in 1.2 - 2.5 fold decrease in milk contamination in comparison with control values, and application of 3 boli a 2-5 fold decrease in both experimental and field studies.
- (3) The direct use of ferrocyn powder resulted in 2.5 to 4.2 fold reductions of radiocaesium in milk.
- (4) Radiocaesium contamination of meat of treated cows was less than that of untreated animals. Three boli per animal resulted in 3-6 fold reductions.
- (5) Applying ferrocyn boli to cattle grazing contaminated land or ferrocyn powder added to the diet resulted in the production of 'clean' milk and meat where previously would not have been possible.
- (6) Application of boli did not influence the clinico-physiological conditions of the treated animals.

## STUDIES PERFORMED IN BELARUS

The Belarus authorities pursued two series of studies. In 1990 and 1991, experiments were conducted on state collective farms to monitor the effect of PB on milk radiocaesium levels, on macro-mineral and trace element concentrations, on body weight, on haematological parameters and on the organoleptic qualities of meat. Between 1992 and 1994, the use of PB was assessed mainly on small farms in the Gomel region.

### RESULTS OF STUDIES CARRIED OUT IN 1990/91

Two sets of experiments are reported. In the first experiment the effects of PB boli on milk radiocaesium levels and on the micromineral composition of milk were measured. The PB used for this experiment was Giese salt produced in Germany. In the second experiment boli with ferrocyan from Ukraine were used to study the effects of PB on radiocaesium content in meat and a long series of physiological and biochemical parameters. All the research and experimental work was organized and conducted by the Research Institute of Agricultural Radiology, Gomel. Under typical conditions of livestock husbandry in Belarus the experiments were performed on grazing milking cows and indoor maintained fattening bull calves.

#### Experiment 1.

In "Rassvet" collective farm (Dobrush area, Gomel region) 18 cows of the black-motley breed weighing 450-500 kg and with a milk yield of 4-6 L/d were divided into three groups: 1st - control (no treatment); the 2nd group were each administered 2 boli and those in the 3rd group - 3 boli. The composition and radiocaesium content of the diet is presented in Table B.33.

TABLE B.33.

| Feeds        | Quantity consumed<br>kg/d | Radiocaesium content |               |
|--------------|---------------------------|----------------------|---------------|
|              |                           | Bq/kg                | Bq/d          |
| Hay          | 3.0                       | 11 100               | 33 300        |
| Cut-grass    | 3.0                       | 211                  | 781           |
| Potatoes     | 8.0                       | 111                  | 412           |
| Concentrates | 3.0                       | 296                  | 888           |
| <b>TOTAL</b> |                           |                      | <b>35 381</b> |

The total activity of a diet was about 35 Kbcq/d. Milk samples were taken at the morning milking and analysed by gamma-spectrometry using an ADKAM 300 (USA). Atomic-absorption spectrophotometry was used to determine macro- and microelements in cows milk on the 8th, 17th and 26th day of the experiment. The total duration of the experiment was 74 days.

Radiocaesium concentration in the milk of the treated animal groups was on average 1.6 - 2.9 times lower than in the milk of untreated animals (Table B.34.).

No significant differences were detected in the macro- and microelement concentrations of milk from control cows and those given 2 or 3 PB boli - Table B.35.

TABLE B.34. THE RADIOCAESIUM CONTENT IN MILK DURING EXPERIMENT ON RASVET COLLECTIVE FARM (Bq/L  $\pm$  SD)

| Animal group       | Initial value | Days after PB application |            |            |            |            |            |            |            |            |            |      |
|--------------------|---------------|---------------------------|------------|------------|------------|------------|------------|------------|------------|------------|------------|------|
|                    |               | 5                         | 8          | 10         | 15         | 19         | 22         | 24         | 27         | 35         | 74         |      |
| Control            | 104           | 257                       | 229        | 230        | 264        | 172        | 196        | 162        | 153        | 127        | 165        |      |
|                    | $\pm 11.1$    | $\pm 44.4$                | $\pm 25.9$ | $\pm 33.3$ | $\pm 22.2$ | $\pm 14.8$ | $\pm 18.5$ | $\pm 14.8$ | $\pm 11.1$ | $\pm 14.8$ | $\pm 14.8$ |      |
| 2 boli             | 141           | 130                       | 84         | 103        | 106        | 94         | 104        | 100        | 81         | 58         | 174        |      |
|                    | $\pm 3.7$     | $\pm 25.9$                | $\pm 11.1$ | $\pm 18.5$ | $\pm 11.1$ | $\pm 14.8$ | $\pm 7.4$  | $\pm 18.5$ | $\pm 7.4$  | $\pm 11.1$ | $\pm 14.8$ |      |
| 3 boli             | 138           | 143                       | 98         | 97         | 91         | 75         | 80         | 75         | 75         | 43         | 104        |      |
|                    | $\pm 14.8$    | $\pm 18.5$                | $\pm 11.1$ | $\pm 18.5$ | $\pm 11.1$ | $\pm 14.8$ | $\pm 11.1$ | $\pm 7.4$  | $\pm 14.8$ | $\pm 11.1$ | $\pm 11.1$ |      |
| Level of reduction | 2 boli        | 0.74                      | 1.99       | 2.73       | 2.23       | 2.50       | 1.84       | 1.89       | 1.61       | 1.88       | 2.18       | 0.95 |
|                    | 3 boli        | 0.76                      | 1.80       | 2.33       | 2.38       | 2.89       | 2.28       | 2.44       | 2.15       | 2.03       | 2.99       | 1.59 |

TABLE B.35. MACRO- AND MICROELEMENT CONCENTRATION IN MILK OF COWS FROM RASSVET COLLECTIVE FARM IN 1990

|         | Date of<br>determination | Name of the element (mg/L milk) |              |             |             |             |              |            |            |
|---------|--------------------------|---------------------------------|--------------|-------------|-------------|-------------|--------------|------------|------------|
|         |                          | Co                              | Cu           | Pb          | Ni          | Li          | Na           | K          | Ca         |
| Control | 8.10.90                  | 0.043±0.003                     | 0.077±0.003  | 0.17±0.01   | 0.055±0.007 | 0.041±0.007 | 1853.7±402.5 | 3697.5±684 | 5746.3±557 |
|         | 17.10.90                 | 0.051±0.002                     | 0.075±0.005  | 0.160±0.008 | 0.270±0.053 | 0.033±0.003 | 1409.6±324   | 2592.7±228 | 5303.9±386 |
|         | 26.10.90                 | 0.060±0.003                     | 0.166±0.085  | 0.160±0.01  | 0.090±0.001 | 0.057±0.007 | 1334.8±157   | 2919.7±353 | 5354.9±484 |
| 2 boli  | 8.10.90                  | 0.045±0.002                     | 0.082±0.0045 | 0.160±0.012 | 0.058±0.004 | 0.095±0.031 | 1823.9±409.6 | 3557.9±585 | 6717.4±944 |
|         | 17.10.90                 | 0.045±0.001                     | 0.110±0.02   | 0.170±0.006 | 0.201±0.07  | 0.072±0.019 | 2031.0±509.5 | 3565.6±633 | 4562.1±402 |
|         | 26.10.90                 | 0.063±0.012                     | 0.712±0.116  | 0.186±0.002 | 0.098±0.005 | 0.071±0.014 | 1823.7±518.3 | 2966.0±552 | 4503.8±564 |
| 3 boli  | 8.10.90                  | 0.044±0.0007                    | 0.061±0.009  | 0.150±0.005 | 0.065±0.007 | 0.049±0.02  | 1951.0±422   | 2954.0±512 | 4339.8±184 |
|         | 17.10.90                 | 0.059±0.003                     | 0.091±0.006  | 0.150±0.008 | 0.068±0.005 | 0.079±0.009 | 2533.1±679   | 2750.3±441 | --         |
|         | 26.10.90                 | 0.056±0.005                     | 0.540±0.12   | 0.210±0.016 | 0.141±0.049 | 0.048±0.009 | 2430.0±441   | 3078.2±607 | 4209.8±604 |
|         |                          | Ba                              | Mg           | Sn          | Sr          | Fe          | Mn           | Zn         |            |
| Control | 8.10.90                  | 6.71±0.53                       | 395.5±230.5  | 1.71±0.09   | 0.088±0.004 | 0.63±0.07   | 0.059±0.04   | 3.43±0.34  |            |
|         | 17.10.90                 | 7.00±0.90                       | 142.1±6.37   | 1.62±0.09   | 0.086±0.005 | 2.33±0.57   | 0.074±0.009  | 4.97±0.41  |            |
|         | 26.10.90                 | 6.20±0.37                       | 186.7±18.2   | 1.65±0.07   | 0.097±0.005 | 0.94±0.10   | 0.060±0.016  | 3.82±0.31  |            |
| 2 boli  | 8.10.90                  | 5.59±0.31                       | 131.0±9.13   | 1.43±0.08   | 0.86±0.004  | 1.45±0.86   | 0.014±0.009  | 3.69±0.58  |            |
|         | 17.10.90                 | 5.90±0.35                       | 186.9±45.9   | 1.48±0.05   | 0.085±0.006 | 1.68±0.69   | 0.067±0.009  | 5.54±0.68  |            |
|         | 26.10.90                 | 5.73±0.18                       | 210.8±10.5   | 1.67±0.03   | 0.090±0.006 | 1.21±0.14   | 0.060±0.005  | 3.13±0.39  |            |
| 3 boli  | 8.10.90                  | 6.15±0.20                       | 250.6±42.9   | 1.53±0.05   | 0.092±0.006 | 1.09±0.52   | 0.130±0.06   | 4.14±0.26  |            |
|         | 17.10.90                 | 6.09±0.41                       | 198.4±8.74   | 1.66±0.11   | 0.088±0.003 | 0.80±0.19   | 0.057±0.003  | 4.38±0.47  |            |
|         | 26.10.90                 | 6.47±0.25                       | 165.0±16.3   | 1.72±0.09   | 0.085±0.001 | 1.83±0.49   | 0.063±0.009  | 3.58±0.36  |            |



## Experiment 2

An experiment on finishing bull-calves maintained indoors was carried out on "Sudkovo" state farm (Choiniki area).

20 'black-motley' bull-calves were divided into 2 groups, 10 animals in each. Two boli per head were administered to the bull-calves of the experimental group. The total radiocaesium activity of the diet was 2.2 Kbkq/d. The diet composition and radiocaesium content in feeds is shown in Table B.36. The duration of the experiment was 88 days.

TABLE B.36. THE DIET COMPOSITION FOR BULL-CALVES AND RADIOCAESIUM INTAKE

| Feeds       | Quantity<br>kg | Radiocaesium content |        |
|-------------|----------------|----------------------|--------|
|             |                | Bq/kg                | Bq/d   |
| Hay         | 2.0            | 447.7                | 895    |
| Grass       | 30.0           | 39.6                 | 11 870 |
| Concentrate | 2.0            | 62.9                 | 1 290  |
| TOTAL       | 34.0           |                      | 22 110 |

At the beginning and at the end of the experiment the following indices were determined in blood, blood plasma and muscle tissue:

- leukocyte count
- erythrocytes, haemoglobin and differentials
- glucose concentration
- amylase activity
- total protein and albumin fractions (albumin, alpha-, beta-, gamma-globulin)
- macro- and microelements in muscle tissue
- spectrometry of meat samples
- physico-chemical indices of muscle tissue.

The experiment showed that the administration of 2 boli reduced the level of contamination in muscle 3.2 times over a three-month period when the daily activity of a ration was 2.2 Kbkq. The specific activity of muscle tissue in untreated and treated calves was 203.5 and 62.9 Bq/kg respectively.

TABLE B.37. BODY WEIGHT (kg) OF BULL-CALVES AFTER BOLUS APPLICATION

| Groups    | Days of experiment |              |
|-----------|--------------------|--------------|
|           | 0                  | 90           |
| Untreated | 245.1 ± 5.7        | 327.6 ± 4.9  |
| Treated   | 241.6 ± 4.8        | 342.5 ± 12.3 |

No negative influence of boli upon the productivity of animals was observed (Table B.37.).

According to the results of organoleptic and chemical tests meat of experimental and control animals complied with the relevant food standard requirements of Belarus (Table B.38.).

Biochemical and haematological blood indices were not significantly different between the treated and untreated groups (Tables B.39. and B.40.).

TABLE B.38. PHYSICO-CHEMICAL INDICES OF MUSCLE TISSUE OF BULL-CALVES AFTER APPLICATION OF FERROCYANIDE BOLI

| Group        | Organoleptic indices |             |                  | Chemical indices                                   |              |                                 |                        |
|--------------|----------------------|-------------|------------------|--|--------------|---------------------------------|------------------------|
|              | Outward appearance   | Consistency | Smell            | Fat  | Acidity (Ph) | Reaction with CuSO <sub>4</sub> | Reaction to peroxidase |
| Control      | Pale pink            | Elastic     | Peculiar to beef | From white colour to yellow.<br>Solid consistency, | 5.5 ± 0.3    | Transparent broth               | Positive               |
| Experimental | Pale pink            | Elastic     | Peculiar to beef | peculiar to fresh beef fat                         | 5.2 ± 0.15   | Transparent broth               | Positive               |

Meat from experimental and control animals met the State standard 23395-78 requirements and methods of physico-chemical examination of meat "Regulations of veterinary-sanitary examination of meat and meat products" and was considered to be fresh and fit for human consumption.

TABLE B.39. DIFFERENTIAL LEUKOCYTE COUNTS IN BLOOD AFTER FERROCYANIDE BOLUS APPLICATION IN CALVES (%)

|           |               |           |           |           |           |            |           |
|-----------|---------------|-----------|-----------|-----------|-----------|------------|-----------|
| Untreated | Initial value | 56.0±5.80 | 3.25±1.90 | 0.5±0.24  | 6.50±1.50 | 33.75±4.60 | —         |
|           | 88            | 63.2±6.01 | 2.00±0.40 | —         | 6.00±1.78 | 28.50±4.00 | 0.25±0.20 |
| Treated   | Initial value | 64.5±6.00 | 4.00±2.00 | 0.75±0.38 | 4.75±1.00 | 24.20±5.00 | —         |
|           | 88            | 64.7±6.00 | 4.10±2.60 | 1.7±1.00  | 7.00±2.00 | 21.70±2.80 | —         |

TABLE B.40. BIOCHEMICAL INDICES OF BLOOD IN BULL-CALVES AFTER APPLICATION OF FERROCYANIDE BOLI

| Groups of animals | Time after application of boli days | Glucose mg/100 ml | Alpha-amylase mg/100 ml | Total protein g/100 ml | Albumin (relative %) | Globulins (relative %) |                |                 |
|-------------------|-------------------------------------|-------------------|-------------------------|------------------------|----------------------|------------------------|----------------|-----------------|
|                   |                                     |                   |                         |                        |                      | Alpha-globulins        | Beta-globulins | Gamma-globulins |
| Untreated         | Initial value                       | 46.8±4.0          | 28.4±6.0                | 7.1±0.1                | 52.0±7.4             | 11.8±2.4               | 22.4±5.4       | 15.0±3.8        |
|                   | 88                                  | 48.0±3.1          | 34.0±7.0                | 6.7±0.2                | 48.9±5.8             | 11.0±1.8               | 23.4±4.7       | 14.6±3.3        |
| Treated           | Initial value                       | 52.8±2.1          | 31.0±4.3                | 6.8±0.4                | 50.0±6.4             | 12.0±3.0               | 24.0±3.8       | 15.4±1.8        |
|                   | 88                                  | 49.4±2.7          | 37.0±3.7                | 6.6±0.1                | 48.7±5.8             | 11.4±2.7               | 22.8±4.0       | 13.7±2.4        |

While analysing micro- and macroelement concentrations in muscle tissues of bull-calves a marked reduction was found in Cu levels in Prussian Blue treated samples (Table B.41.). This requires further study as this reduction was not observed in milk from cows administered boli.

TABLE B.41. CONTENT OF MACRO- AND MICROELEMENTS IN MUSCLE TISSUE OF BULL-CALVES AFTER APPLICATION OF FERROCYNE BOLI (mg/kg)

| Name of macro-<br>or microelements | Group           |                           |
|------------------------------------|-----------------|---------------------------|
|                                    | Control         | Treated                   |
| K                                  | 5680.5 ± 2380.0 | 4804.0 ± 301.4            |
| Ca                                 | 1801.7 ± 714.8  | 1914.6 ± 840.2            |
| Na                                 | 1438.3 ± 280.0  | 1401.3 ± 184.3            |
| Co                                 | 0.082 ± 0.001   | 0.079 ± 0.04              |
| Cu                                 | 0.560 ± 0.14    | 0.071 ± 0.01 <sup>a</sup> |
| Pb                                 | 0.334 ± 0.10    | 0.304 ± 0.05              |
| Ni                                 | 0.107 ± 0.01    | 0.128 ± 0.03              |
| Li                                 | 0.124 ± 0.03    | 0.178 ± 0.02              |
| Mg                                 | 239.7 ± 30.1    | 241.6 ± 18.4              |
| Ba                                 | 0.801 ± 0.14    | 0.794 ± 0.15              |
| Mo                                 | 0.810 ± 0.06    | 0.879 ± 0.11              |
| Sn                                 | 1.890 ± 0.23    | 1.887 ± 0.20              |
| Zn                                 | 40.7 ± 6.0      | 40.7 ± 7.8                |
| Fe                                 | 16.6 ± 3.0      | 17.4 ± 0.3                |
| Cd                                 | 0.024 ± 0.003   | 0.021 ± 0.004             |
| Cr                                 | 0.218 ± 0.07    | 0.210 ± 0.02              |
| Mn                                 | 0.083 ± 0.004   | 0.092 ± 0.001             |

<sup>a</sup> - p < 0.05 - as compared with control

#### RESULTS OF FIELD TRIALS CARRIED OUT IN 1992-1994

These trials were carried out on private farms in the Lelchitsy and Elsk districts of the Gomel region. A special feature of the Lelchitsy district is that more than 70% of its territory is highly afforested and boggy. This hampers the use of common countermeasure strategies. Nevertheless, within the Lelchitsy district an area of 953 ha of seeded pastures has already been allotted to the private sector and 1 227 ha more are planned to be regrassed. In 1994, 5 296 cows were owned by private farm holders in the district in 1-3 cow herds. For each cow this was, on average, 0.17 ha of seeded pasture available, i.e. less than 50% of the area required.

In the Lelchitsy District milk yield of the local black-and-white breed is ca. 8-10 litres per day or 2 500 litres per lactation. Live weight gain of younger animals is 350-500 g/day. Mature live weight of local cows varies from 350 to 550 kg.

After the time of the trial, <sup>137</sup>Cs contamination density of the land used for grazing and hay making in the localities of the Lelchitsy District was not high; it ranged from 1.5 to 8 Ci/km<sup>2</sup>.

The contamination level in grass, forage, hay, feeds and milk in each location was measured at the beginning of the study.

**TABLE B.42. Cs-137 CONTAMINATION LEVELS OF FEEDS AND MILK (Bq/kg) IN 1992**

| Lelchitsy District | Grass         | Milk <sup>a</sup> |
|--------------------|---------------|-------------------|
| 1) Derzhinsk       | 821 - 2 261   | 145 - 2 317       |
| 2) Borovoe         | 1 029 - 1 389 | 314 - 435         |
| 3) Tartak          |               | 217 - 376         |
| 4) Slobodka        | 439 - 1 049   | 268 - 363         |
| 5) Markovskoe      | 389 - 1 761   | 196 - 512         |
| 6) Kortanichi      |               | 222 - 596         |
| 7) Kalinovka       |               | 235 - 1 305       |
| 8) Krasnoberezhye  | 2 491         | 197 - 460         |
| 9) Manchitsa       | 1 179 - 2 830 | 168 - 483         |
| 10) Zabolotye      | 3 372         | 70 - 453          |
| 11) Rubezh         |               | 120 - 385         |
| 12) Lelchitsy      | 1 676         | 122 - 505         |
| 13) Glushkovichi   | 2 298         | 214 - 388         |
| 14) Milashevichi   |               | 132 - 235         |
| 15) Pribylovichi   | 1 233         | 228 - 370         |
| 16) Lisnoe         |               | 333 - 550         |
| 17) Krupka         |               | 196 - 347         |
| 18) Zarubanoe      |               | 112 - 349         |
| 19) Mekhach        |               | 120 - 420         |
| 20) Simonichskaya  |               | 210 - 308         |

<sup>a</sup> Temporary Permissible Limit for milk in 1992 was 185 Bq/L

Prussian Blue was given in one of three ways: in a bolus, - 3 boli per cow each 6 weeks in grazing season if the level of <sup>137</sup>Cs was lower than 300 Bq/L; in 5 kg weight salt-licks which lasted 90-100 days; as compounded concentrate (0.6% PB, 0.5 kg concentrate/day) during lactation.

The results of bolus treatment in a herd of milking cattle are listed in Table B.43.

TABLE B.43. Cs-137 CONTENT IN MILK DURING BOLUS TREATMENT OF ANIMALS IN THE LELCHITSY REGION (n = 5 ANIMALS PER GROUP)

| Groups of animals | Cs-137 content in milk, Bq/L |                |                |                |                |  |
|-------------------|------------------------------|----------------|----------------|----------------|----------------|--|
|                   | Before                       | 10 days        | 20 days        | 30 days        | 40 days        |  |
|                   |                              | Derzhinsk      |                | First herd     |                |  |
| Control           | 2 530<br>± 202               | 2 835<br>± 181 | 1 833<br>± 302 | 1 854<br>± 234 | 1 931<br>± 370 |  |
| Treated           | 1 779<br>± 429               | 848<br>± 96    | 323<br>± 37    | 257<br>± 43    | 434<br>± 171   |  |
| Reduction         | -                            | 3.3            | 5.6            | 7.2            | 4.4            |  |
|                   |                              | Derzhinsk      |                | Second herd    |                |  |
| Control           | 1 510<br>± 116               | 1 397<br>± 251 | 911<br>± 182   | 1 078<br>± 182 | 1 721<br>± 147 |  |
| Treated           | 1 062<br>± 147               | 270<br>± 42    | 369<br>± 128   | 244<br>± 55    | 226<br>± 104   |  |
| Reduction         | -                            | 5.2            | 2.5            | 4.4            | 7.6            |  |
|                   |                              | Slobodka       |                |                |                |  |
| Control           | 269<br>± 8                   | 305<br>± 29    | 288<br>± 24    | 277<br>± 18    | 298<br>± 19    |  |
| Treated           | 356<br>± 18                  | 189<br>± 36    | 126<br>± 22    | 115<br>± 25    | 113<br>± 17    |  |
| Reduction         | -                            | 1.6            | 2.3            | 2.4            | 2.6            |  |

Note: Each animal was treated with 3 boli

The effect of Prussian Blue material on <sup>137</sup>Cs reduction in milk, where levels of contamination were low, was also studied (Table B.44.).

TABLE B.44. THE CHANGE IN Cs-137 CONCENTRATION IN MILK WHEN THE PRUSSIAN BLUE CONTAINING BOLI WERE USED AT LOW LEVELS OF Cs-137 ON THE FARM "DUBOVY LOG" (DOBRUSH DISTRICT), Bq/L

| Groups of animals | Time of milk sampling (days after application) |           |           |           |           |
|-------------------|--|-----------|-----------|-----------|-----------|
|                   | 0  | 5         | 10        | 15        | 20        |
| Control<br>n = 5  | 33<br>± 2                                      | 22<br>± 2 | 19<br>± 2 | 36<br>± 2 | 45<br>± 1 |
| Treated<br>n = 4  | 29<br>± 3                                      | 13<br>± 1 | 14<br>± 1 | 15<br>± 3 | 15<br>± 3 |

Note: Each animal was given 3 boli

The data shown in Table B.44. indicate that Prussian Blue is effective even at low milk <sup>137</sup>Cs concentrations.

The efficiency of treatment with salt-licks and Prussian Blue-containing mixed feeds is shown in Tables B.45. and B.46.

TABLE B.45. THE EFFECT OF OFFERING PB CONTAINING SALT-LICKS ON CS-137 LEVELS IN MILK (Bq/L) - LELCHITSY DISTRICT

| Locality  | Animal group | PB content in salt-lick (%) | Days after exposure to salt licks |                |                |                |                |
|-----------|--------------|-----------------------------|-----------------------------------|----------------|----------------|----------------|----------------|
|           |              |                             | 0                                 | 10             | 20             | 30             | 40             |
| Derzhinsk | 1            | 3                           | 2 449<br>± 170                    | -<br>-         | 762<br>± 122   | 348<br>± 141   | 614<br>± 325   |
|           | 2            | 10                          | 1 365<br>± 130                    | 644<br>± 174   | 566<br>± 178   | 325<br>± 67    | 910<br>± 248   |
|           | Control      | -                           | 2 531<br>± 200                    | 2 812<br>± 178 | 1 813<br>± 303 | 1 857<br>± 233 | 1 931<br>± 344 |

For the period of the experiment, the <sup>137</sup>Cs content in milk was reduced 1-4 fold in the first group of cows and by 2-4 fold in the second group.

TABLE B.46. THE EFFECT OF GIVING PRUSSIAN BLUE-CONTAINING MIXED FEEDS ON Cs-137 LEVELS IN MILK ON THE COLLECTIVE FARM "NA VARTE" (LELCHITSY DISTRICT) - Bq/L

| Group of cows     | Days after exposure to PB containing feed |           |           |           |
|-------------------|---|-----------|-----------|-----------|
|                   | 0   | 6         | 16        | 26        |
| Treated<br>n = 20 | 45<br>± 4                                 | 19<br>± 3 | 18<br>± 3 | 17<br>± 2 |
| Control<br>n = 10 | 44<br>± 7                                 | 33<br>± 4 | 48<br>± 7 | 35<br>± 6 |

The inclusion of Prussian Blue into finishing rations during the last 30 days before slaughter resulted in a 2-3-fold reduction in <sup>137</sup>Cs content in the muscle tissues of treated animals as compared with that of the control group.

The results of long-term field studies showed that under conditions of radioactive contamination the use of Prussian Blue materials is a reliable method of reducing <sup>137</sup>Cs content in milk when the application of traditional countermeasures is impracticable in the soil-plant chain. In Belarus, the production of Prussian Blue-containing mixed feeds, boli and salt-licks is well established. This enables the country to meet the demand of private and collective farmers for these products.

Treatment of cows in the private sector with Prussian Blue materials systematically reduces <sup>137</sup>Cs level in milk by a factor of 2-3. As a result, the internal dose in man can be reduced by 25-30% (assuming that the dose from milk is no less than 50-60% of the internal radiation dose).



## The effect of manure from animals treated with ferrocyn-containing boli on the soil-plant transfer of radiocaesium

In 1992-1993, the influence of cattle manure from animals treated with ferrocyn containing boli on the transfer of  $^{137}\text{Cs}$  from soil into potatoes was studied (Table B.47.). The experiment was performed in the Vetka District (Gomel Region). The soil is podzolic, sandy, Ph is 3.9,  $\text{P}_2\text{O}_5$  is 23.2 mg/100 g,  $\text{K}_2\text{O}$  is 10.1 mg/100 g,  $\text{CaO}$  is 16.8 mg/100 g of soil.  $^{137}\text{Cs}$  contamination density was 393 K $\text{Bq}/\text{m}^2$  (ca. 11 Ci/ $\text{km}^2$ ). For each square meter, 3 g Prussian Blue material was applied to the manure. The application rate of the manure resulted in the spread of 3 g/ $\text{m}^2$ .

TABLE B.47. Cs-137 CONTENT IN POTATOES AFTER APPLICATION OF MANURE DRESSED WITH PB (Bq/kg)

| Test variants  | 1992   | 1993   |
|--|--------|--------|
| Control $\text{N}_{90}\text{P}_{100}\text{K}_{140}$<br>+ 50 ton/ha of manure                       | 24 ± 7 | 13 ± 2 |
| $\text{N}_{90}\text{P}_{100}\text{K}_{140}$ + 50 ton/ha of<br>manure dressed with Prussian<br>Blue | 18 ± 1 | 15 ± 3 |

As can be seen there was no significant effect of Prussian Blue addition to cattle manure on the  $^{137}\text{Cs}$  content of potatoes grown in contaminated soil.

In 1994, in the vicinity of the village of Savichi located in the evacuation zone, an experiment was carried out on the application of Prussian Blue to the sod cover of a native meadow at a rate of 20 kg/ha followed by ploughing, harrowing and seeding with ryegrass (Table B.48.). The soil is soddy cryptopodzolic, gley, loamy, Ph of 3.7,  $\text{P}_2\text{O}_5$  of 4.0 mg/100 g,  $\text{K}_2\text{O}$  of 9.8 mg/100 g, Ca of 39.0 mg/100 g of soil.  $^{137}\text{Cs}$  contamination density was 982 K $\text{Bq}/\text{m}^2$  (ca. 23 Ci/ $\text{km}^2$ ).

TABLE B.48. THE INFLUENCE OF PRUSSIAN BLUE ON Cs-137 CONTENT IN RYEGRASS (Bq/kg AIR-DRIED WEIGHT)

| Variants   | 1 repl. | 2 repl. | 3 repl. | 4 repl. | Mean (sd)   |
|--|---------|---------|---------|---------|-------------|
| $\text{N}_{60}\text{P}_{90}\text{K}_{120}$ + 8 t/ha<br>of lime - I | 2 280   | 2 150   | 2 150   | 3 880   | 2 615 (845) |
| I + 2 g/ $\text{m}^2$ of<br>Prussian Blue                          | 2 100   | 1 225   | 2 370   | 2 810   | 2 125 (670) |

The results of the experimental studies demonstrated that the application of 20 kg of Prussian Blue/ha (which corresponds to 330 tons of Prussian Blue manure produced by animals consuming 3 g of PB/animal/day) did not significantly reduce the uptake of  $^{137}\text{Cs}$  by ryegrass.

## CONCLUSION OF STUDIES

On the whole, villagers treat their animals with PB containing materials such as boli and salt-licks, etc. if public opinion formers do so. The opinions of local administrators and specialists are important for the residents living in contaminated areas. If state farm managers, animal production specialists, veterinarians or medical workers readily treat their own animals with boli, salt-licks and mixed feeds the rest of the residents usually follow their example. However, a cow owner's first

impression of the procedure of bolus treatment is a poor one. So treatment with Prussian Blue-containing mixed feeds and salt-licks is preferable. In the sphere of social status the ability of a small farmer to produce clean produce is a major motive. In general, managers of large herds also routinely use Prussian Blue-containing preparations which is testament to their effectiveness.

## ANNEX C

### TOXICOLOGICAL CONSIDERATIONS

Work carried out during the 1960s demonstrated the effectiveness of Prussian Blue as an agent to bind radiocaesium. This work was initiated to develop countermeasures to reduce or prevent the radiocaesium contamination of animal-derived foods as a consequence of weapons-test fallout or enhance its elimination from the animal's body (see Giese, 1987a, 1988, 1989).

Following the Chernobyl reactor accident interest in Prussian Blue compounds was restimulated and studies have been carried out since 1986 in many countries affected by Chernobyl fallout.

Prussian Blue, which is synonymous with Iron Blue or Berlin Blue, is a name given to a number of ferric hexacyanoferrates; perhaps the most intensively studied form of Prussian Blue following the Chernobyl accident has been ammonium ferric cyanoferrate (AFCF - Giese salt).

Investigations have been carried out in a number of countries, including Austria, France, Germany, Greece, Hungary, Norway, Belarus, Russian Federation, Ukraine and the United Kingdom, on the use of Prussian Blue as a countermeasure to reduce radiocaesium levels in a wide range of animals (cows, sheep, pigs, goats, deer, domestic fowl) and their products.

An important consideration in the practical use of Prussian Blue compounds in agriculture is their possible toxicity. This is similarly of great importance when such compounds are used to enhance the clearance of radiocaesium from accidentally contaminated humans. A number of studies have, therefore, examined their chemical stability and toxicity. Such investigations have been undertaken with laboratory rodents, domesticated animals and humans. Other studies relating to the use of Prussian Blue in thallotoxicosis and its use as a food additive are also relevant to any toxicological effects of such compounds. The work carried in these areas is reviewed below.

#### Studies relevant to any toxic effects of Prussian Blue in laboratory rodents

The studies carried out by Nigrovic (1963; 1965) and Nigrovic et al. (1966) on the effects of Prussian Blue on caesium metabolism in the rat also included considerations of any toxic effects. It was shown that ferric cyanoferrate ( $\text{Fe}_4[\text{Fe}(\text{CN})_6]_3$ ) prevented the tissue uptake of orally administered  $^{137}\text{Cs}$ , enhanced its clearance from the body and also had no toxic side effects. In these studies doses of 100 mg Prussian Blue/day for 3 days (Nigrovic, 1963) or 100 mg/day for 11 days (Nigrovic, 1965) were administered. These studies were extended by Nigrovic et al. (1966) who showed that there were no toxic side effects and no impairment of growth in young rats maintained for 120 days on a diet containing 1% ferric cyanoferrate.

The chronic consumption of ferric cyanoferrate was also investigated by Richmond and Bunde (1966). These workers provided Prussian Blue over a 60 day period. No toxic effects of Prussian Blue ingestion were observed.

The metabolic behaviour of ferric hexacyanoferrate was investigated by Dvorak et al. (1971). Following intravenous injection they noted that the  $[\text{Fe}(\text{CN})_6]^{4-}$  ion was rapidly and virtually completely excreted via the urine. Following oral administration they reported that a small amount of the hexacyanoferrate ion was absorbed but there was no evidence for its decomposition and also no toxic side effects were observed when 2% ferric hexacyanoferrate was provided in the drinking water of rats over a 12 week period.

As a result of studies on the effects of various metal ferrocyanides bound in an anion exchange resin in removing radiocaesium from sea water, milk and fuel processing waste, Inuma et al. (1971) examined the effectiveness of a nickel ferrocyanide/resin complex (NiFC)R on the

elimination of  $^{137}\text{Cs}$  from rats and mice. They also examined any toxic effects as a forerunner to human studies. A diet containing 2% (NiFC)R was provided to weaning rats for 152 days when they were slaughtered and histopathological studies carried out on the livers, kidneys, spleens, thymus glands, brains, testes and femurs. No histopathological changes were noted.

Kostial et al. (1981) examined the effects of a composite treatment consisting of calcium alginate, ferric cyanoferrate and potassium iodide on the retention of  $^{90}\text{Sr}$ ,  $^{137}\text{Cs}$  and  $^{131}\text{I}$  in rats. The ferric cyanoferrate represented 2.5% of the diet and the experimental period was 4 weeks. In this study a large number of parameters were examined to determine any toxic effects of the treatment. The haematological parameters included packed cell volume, haemoglobin concentration, mean corpuscular volume, mean corpuscular haemoglobin content and leukocyte counts. Samples of liver, kidney and femur were assayed for iron, zinc and manganese. The mineral, water and organic components of the femurs were determined and morphometric measurements on the femurs were also carried out. Histopathological examinations were made on the lung, heart, liver, spleen, stomach, duodenum, jejunum, ileum, colon, kidneys, thyroid and trachea of the rats. The results showed the mixture to be practically non toxic. There was a slight reduction in the haemoglobin content and liver iron in treated animals. These were the only effects seen and were ascribed to the alginate component of the mixture.

Kargacin et al. (1985) carried out studies with a mixture of similar composition but, in addition, included Ca-DTPA and Zn-DTPA (as antidotes for transuranic elements). Any effects on the health and well being of the rats was assessed by evaluating bodyweight, haematological and histopathological examinations and trace element analysis. A number of slight effects were seen (slightly lower erythrocyte number, slightly lower haemoglobin and haematocrit values and very mild histopathological changes in the kidney). These changes are consistent with the known effects of alginate and Ca-DTPA and hence no toxic effects due to Prussian Blue were observed.

Further evidence of the non-toxicity of Prussian Blue, in the form of ammonium ferric cyanoferrate (AFCF) was shown by the observation that its  $\text{LD}_{50}$  in mice was well above 5 g AFCF/kg bodyweight (Sterner and Chibanguza, 1987; see Giese, 1988).

There is one study reported in the literature that suggests that sodium ferrocyanide causes tissue changes in the rat (Anon, 1969). In this study diets containing 0, 0.05, 0.5 and 5.0% sodium ferrocyanide were fed to rats for 90 days. The 5% sodium ferrocyanide diet depressed growth rate in male rats but not females and also reduced the haemoglobin concentrations and haematocrit values in both sexes. All other haematological indices were normal. Kidney enlargement occurred on the 5% ferrocyanide diet and a number of histological changes in the kidney were reported on the 0.5% and 5.0% diets. The report concluded that the 0.05% concentration of sodium ferrocyanide was without effect. However, this report represented a summary of a previously unpublished study carried out in 1959. Since it is not an original report giving actual data it may be prudent to treat the interpretation with caution; to validate the reported observations would require the study to be repeated. In addition, it should be noted that sodium ferrocyanide is an approved food additive for use in the human diet (see later section).

#### **Study with dogs**

Madshus et al. (1966) administered doses of 1.5 and 3.0 g ferric cyanoferrate (11) to 3 month old Alsatian dogs for a period of 10 days. The body weights and general well being of the dogs was unaffected by the Prussian Blue.

## **Prussian Blue toxicity studies in ruminants**

### **(a) Cattle**

Studies of the metabolism of the AFCF form of Prussian Blue by dairy cows were carried out by Arnaud et al. (1988) using [<sup>14</sup>C] labelled material. When 3 g AF<sup>14</sup>CF were orally administered nearly 100% of the radioactivity ( $95 \pm 5\%$  and  $91 \pm 7\%$  in the two experimental animals) were recovered in the faeces over a period of 9 days. Total cumulative radioactivity in the urine and milk over this 9 day period was 0.19 - 0.47% and 0.068 - 0.071%, respectively, and very low levels appeared in the plasma, blood cells, expired CO<sub>2</sub> and various tissues (pancreas, heart, liver, spleen and muscle). The tissue levels were not greater than 6 Bq/kg above background. The authors considered that the radioactivity in milk was due to thiocyanate. Arnaud et al. (1988) concluded that AFCF constitutes a safe food additive to prevent the transfer of dietary radiocaesium to milk.

It was similarly shown that when 2 lactating cows were dosed with 100  $\mu$ Ci [<sup>59</sup>Fe] AFCF for 15 days the total faecal excretion amounted to 102.2% and 100.6% in those 2 animals (Giese, 1987b, 1988).

Overdosing of lactating cows (20 g AFCF/day for 15 days) revealed no histopathological changes in 11 different tissues per animal and no changes in bacterial and protozoal counts in rumen contents (Giese et al., 1987). The milk of these cows was analysed for free cyanides. None could be detected (detection limit: 0.1  $\mu$ g CN/g milk). In any case if cyanides had been liberated from AFCF by digestive processes the majority of it would have been detoxified to thiocyanate (SCN<sup>-</sup>) and thiocyanate is normally found in milk, plasma and urine. To check this point thiocyanate concentrations in plasma and milk of overdosed cows were compared with those of controls. No significant differences in thiocyanates were detected.

### **(b) Sheep**

The provision of 2 g/day Prussian Blue (in the form of AFCF) in the diet of lactating ewes did not produce any adverse effects on behaviour, food intake, bodyweight or milk production over an experimental period of 90-100 days (Daburon et al., 1991). Similarly the provision of 1% ferric ferrocyanide in the drinking water of lactating ewes over a period of 23 days had no effect on bodyweight (Ioannides et al., 1991).

Histopathological studies were carried out in sheep which had received 5 g AFCF/day for 15 days. No histopathological changes were seen in the 11 different tissues examined (Giese et al., 1987). In addition, no physiological changes in Na<sup>+</sup>, Cl<sup>-</sup> and phosphate transport through the rumen epithelium and no changes in the rumen bacterial and protozoal counts were seen.

A number of studies have been carried out to examine any toxicological effects of indwelling rumen boli containing Prussian Blue (AFCF form); such boli provide a slow release mechanism for AFCF. In the work of Pearce et al. (1989) boli containing AFCF (20%), stone meal and saturated fatty acids (as binder) were used. The duration of the study was 7 weeks and during this period the animals were regularly examined by an experienced veterinarian and blood samples were taken for the assessment of a range of biochemical and haematological parameters; these clinical examinations and analyses showed that the Prussian Blue boli did not affect the health and well being of the animals.

Histopathological studies were also carried out to investigate any effects of such boli (Pearce and Smyth, in prep.). Four weeks after the administration of AFCF-containing boli or continuous daily dosing with particulate AFCF (via gelatin capsules) the sheep were slaughtered and histopathological examinations carried out on buccal mucosa, rumen, reticulum, omasum, abomasum, jejunum, ileum, colon, leg muscle, spleen, liver, heart, kidney, lungs and brain. No differences were seen between the sheep which received boli and the untreated control animals.

Subsequent animal house and field studies (Pearce et al., in prep.) and field studies (Pearce et al., unpublished observations) were carried out using a bolus consisting of AFCF (15%), stone meal and beeswax (as binder) over periods of 7 to 13 weeks and similar clinical veterinary examinations and blood biochemical and haematological examinations were carried out. The blood biochemical profile assays included total protein albumin, globulin,  $\text{Ca}^{++}$ ,  $\text{Mg}^{++}$ ,  $\text{Cu}^{++}$ , phosphate, urea, creatinine,  $\beta$  hydroxybutyrate, glutathione peroxidase and creatine phosphokinase. The haematological parameters were packed cell volume, haemoglobin, red cell count, white cell count, white cell volume, mean cell volume, mean corpuscular haemoglobin, mean corpuscular haemoglobin concentration and differential white cell counts (as a percentage of the total white cell count). In all cases the values obtained were within the normal ranges for these parameters and there were no differences between bolus treated and control sheep. These data reconfirm the previous studies that the AFCF-containing boli did not affect the health and well-being of the animals. Hansen et al. (in prep.) also observed that AFCF-containing boli had no effect on the body-weight of lambs, again indicating no detrimental effect on the health and digestive processes of sheep.

### **The effects of the Prussian Blue when administered to humans**

The studies by Nigrovic and co-workers (Nigrovic, 1963; Nigrovic, 1965; Nigrovic et al., 1966) which demonstrated the high efficiency of Prussian Blue in eliminating radiocaesium in rats were extended by the same group to humans (Madshus et al., 1966; Madshus and Strömme, 1968). In the former study the human subjects ingested 3 g Prussian Blue daily; although the duration of Prussian Blue ingestion is not clearly stated the data presented suggests the time period was 20 days. The biological half life of previously administered  $^{137}\text{Cs}$  was reduced by 60-70% and no side effects were noted except for slight constipation. In particular it is noteworthy that there was no change in the  $^{40}\text{K}$  content of the body. This work was further pursued by Madshus and Strömme (1968) using 6 human volunteers who received 1.5 or 3.0 g Prussian Blue/day and similar results were obtained. There was no observable prophylactic effect. Madshus et al. (1966) suggested the use of Prussian Blue as the treatment of choice in humans for cases where radiocaesium had been ingested.

As outlined in the section above on laboratory rodents Inuma et al. (1971) showed the effectiveness of nickel ferrocyanide-anion exchange resin (NiFC)R in enhancing the elimination of  $^{137}\text{Cs}$  from rats. These authors extended the work to examine the effects of (NiFC)R in 2 human volunteers who had ingested  $^{137}\text{Cs}$ . Various doses of (NiFC)R were used up to a maximum of 2.2 g/d for 9 days. The (NiFC)R increased the excretion of  $^{137}\text{Cs}$  but had no side effects on the 2 subjects during or after the experimental period. The potassium content of the body was unaffected.

Prussian Blue (ferric cyanoferrate,  $\text{Fe}_4 [\text{Fe}(\text{CN})_6]_3$ ) was administered to individuals contaminated with  $^{137}\text{Cs}$  in the Goiania accident in Brazil in September 1987 (Roberts, 1987; Lipsztein et al., 1991). Doses ranging from 1 to 10 g/day were administered during the first month after the accident although the actual duration of administration is not specified in these reports. The Prussian Blue was effective in reducing the  $^{137}\text{Cs}$  contamination; there was no mention of the presence or absence of any other effects.

Prussian Blue was also used as a therapeutic treatment to enhance the excretion of radiocaesium by Chinese students who were internally contaminated with radionuclides released from Chernobyl, while staying in Sofia and Profdef, Bulgaria (Tang et al., 1988). The dose given was 3 g/day for 6 days.

### **The use of Prussian Blue in the treatment of thallotoxicosis in humans**

Thallium salts are still widely used as rodenticides in many countries despite the fact that thallium poisoning (thallotoxicosis) is a potential hazard due to accidental or intentional ingestion by humans (Heydlauf, 1969; Moeschlin, 1980; Thompson, 1981; Forth, 1983). Hence, it represents a serious health problem in all those countries where the sale and use of thallium-containing preparations is unrestricted.

Many antidotes have been suggested and Heydlauf (1969) showed the effectiveness of Prussian Blue. Prussian Blue therapy was also strongly recommended by others (Kamerbeek et al., 1971; Moeschlin, 1980; Thompson, 1981; Forth, 1983; Hartvig, 1984).

Numerous experimental studies have been carried out on thallium toxicity in the rat and Prussian Blue was generally shown to be the most effective agent for enhancing the excretion of thallium (Rauws, 1974; Lehman and Favari, 1985; Leloux et al., 1990). In all these studies the potassium ferric cyanoferrate (11) form of Prussian Blue was used at a rate of 100 mg/kg body weight/day for 3 days (Leloux et al., 1990), 8 days (Lehman and Favari, 1985) or 3 to 6 days (Rauws, 1974).

Prussian Blue may be considered to be the best antidote because it interferes with the entero-systemic circulation of thallium and enhances its faecal excretion. It is especially noteworthy that, since 1971, Prussian Blue therapy for thallium intoxication has been recommended by the National Poison Control Center of the National Institute of Public Health in the Netherlands (Van Kesteren et al., 1980).

In this context a recent report of the use of Prussian Blue to treat 9 patients suffering from thallium poisoning is pertinent (Pai, 1987). These individuals received a dose of 2 g Prussian Blue (potassium ferric hexacyanoferrate 11) three times a day for a total of 6 weeks. This represented an intake of 252 g Prussian Blue to patients ranging from 16-70 years, including one pregnant woman. Sixteen weeks after the initial poisoning all had returned to active life and no side effects were seen during Prussian Blue therapy.

#### **Uses of ferrocyanide compounds in relation to the human diet**

It is very pertinent to any consideration of the possible toxic effects of Prussian Blue in the human to note that sodium ferrocyanide is used as an anti-caking agent in common table salt at levels up to 15 ppm (Anon, 1969). Calcium and potassium ferrocyanides are also used as anti-caking agents (World Health Organization, 1974).

Moreover, hexacyanoferrate compounds find uses as precipitants for removing metal ions from wine. Tetrapotassium hexacyanoferrate 11 ( $K_4[Fe(CN)_6]$ ) and ferrous potassium hexacyanoferrate 11 ( $K_2Fe[Fe(CN)_6]$ ) are used to eliminate copper and iron in wines in Europe and the USA (Anon, 1969; Wencker et al., 1989). It has similarly been shown that these treatments are effective in removing cadmium from wine (Wencker et al., 1990).

#### **CONCLUSIONS**

The studies described above indicate that Prussian Blue compounds are an effective countermeasure for radiocaesium. They had no adverse effects on animal health and production and similarly no toxic effects were noted in humans when Prussian Blue was used experimentally or therapeutically. The consumption of products from Prussian Blue treated animals would not be expected to have any effects on human health and it is pertinent to note that on the basis of the work carried out with AFCF this compound has been officially approved for use with domestic animals in Germany, Austria and Norway since 1987.

## BIBLIOGRAPHY

ANON., Short-term feeding study of sodium ferrocyanide in rats, *Food Cosmet. Toxicol.* **7** (1969) 409-410.

ARNAUD, M.M., CLEMENT, C., GETAZ, F., TANNHAUSER, F., SCHOENEGGE, R., BLUM, J., GIESE, W., Synthesis, effectiveness and metabolic fate in cows of the caesium complexing compound ammonium-ferric-hexacyanoferrate labelled with  $^{14}\text{C}$ , *Journal of Dairy Research* **55** (1988) 1-13.

DABURON, F., ARCHIMBAUD, Y., COUSI, J., FAYART, G., HOFFSCHIR, D., CHEVELLEREAU, I., LE CREFF, H., Radiocaesium transfer to ewes fed contaminated hay after the Chernobyl accident: Effect of vermiculite and AFCF (ammonium ferricyanoferrate) as countermeasures, *Journal of Environmental Radioactivity* **14** (1991) 73-84.

DVORAK, P., GUNTHER, M., ZORN, V., CATSCH, A., Metabolisches Verhalten von kolloidalem Ferricyanoferrate (11), *Naunyn-Schmiedeberg's Archives of Pharmacology* **269** (1971) 48-56.

FORTH, W., Thallium-Vergiftung, *Münch. med. Wschr.* **125** (1983) 45-50.

GIESE, W., Der fütterungsabhängige Radio-cäsium - Expositionspfad für nutzbare Haustiere sowie Möglichkeiten zur Verminderung der radioaktiven Strahlenbelastung, *Übers. Tierernährung* **15** (1987a) 113-134.

GIESE, W., Über den Grad der Absorbierbarkeit von Fe-59 aus Ammonium-Eisen-59-Hexacyanoferrat durch Milchkühe, 2. Teilbericht (B), Fachgebiet Medizinische Physik, Tierärztliche Hochschule Hannover (1987b).

GIESE, W.W., Ammoniumferric-cyano-ferrate (11) (AFCF) as an effective antidote against radiocaesium burdens in domestic animals and animal derived foods, *British Veterinary Journal* **144** (1988) 363-369.

GIESE, W.W., Countermeasures for reducing the transfer of radiocaesium to animal derived foods, *The Science of the Total Environment* **85** (1989) 317-327.

GIESE, W., MOLNER, S., LINGENS, K., Unpublished data cited in Giese, W. (1988), *British Veterinary Journal* **144** (1987) 363-369.

HARTVIG, P., Chemical principles of chelate therapy in neurotoxicology, *Acta Neurol. Scand.* **70** (suppl. 100) (1984) 99-202.

HEYDLAUF, H., Ferric-cyanoferrate (11): an effective antidote in thallium poisoning, *European Journal of Pharmacology* **6** (1969) 340-344.

HANSEN, H.S., HOVE, K., BARVIK, K., STRAND, P., SELNES, T. (in preparation).

HOVE, K., STRAND, P., SALBU, B., OUGHTON, D., ASTASHEVA, N., VASILIEV, A., RATNIKOV, A., AVERIN, V., FIRSAKOVA, S., CRICK, M., RICHARDS, J.I., "Use of caesium binders to reduce radiocaesium contamination of milk and meat in Belarus, Russia and Ukraine", *Environmental Input of Radioactive Releases (Proc. Int. Symp. Vienna, 1995)*, IAEA, Vienna (1995).

IINUMA, T.A., IZAWA, M., WATARI, K., ENOMOTO, Y., MATSUSAKA, N., INABA, J., KASUGA, T., NAGAI, T., Application of metal ferrocyanide-anion exchange resin to the enhancement of elimination of  $^{137}\text{Cs}$  from human body, *Health Physics* **20** (1971) 11-21.



- IOANNIDES, K.G., MANTZIOS, A.S., PAPPAS, C.P., Influence of Prussian Blue in reducing transfer of radiocaesium into ovine milk, *Health Physics* **60** (1991) 262-264.
- KAMERBEEK, H.H., RAUWS, A.G., TEN HAM, M., VAN HEIJST, A.N.P., Prussian Blue in therapy of thallotoxicosis, *Acta Med. Scand.* **180** (1971) 321-324.
- KARGACIN, B., MALJKOVIC, T., BLAUNSA, M., KOSTIAL, K., The influence of a composite treatment for internal contamination by several radionuclides on certain health parameters in rats, *Arh. hig. rada toksicol.* **36** (1985) 165-172.
- KOSTIAL, K., KARGACIN, B., RABAS, I., BLANUSA, M., MALJKOVIC, T., MALKOVIC, V., CIGANOVIC, M., Simultaneous reduction of radioactive strontium, caesium and iodine retention by single treatment in rats, *The Science of the Total Environment* **22** (1981) 1-10.
- LEHMAN, P.A., FAVARI, L., Acute thallium intoxication: Kinetic study of the relative efficacy of several antidotal treatments in rats, *Archives of Toxicology* **57** (1985) 56-60.
- LELOUX, M.S. NGUYEN PHU LICH, CLAUDE, J.R., Experimental studies of several antidotal treatments on the tissue distribution and elimination of thallium, after subacute intoxication, *Journal de Toxicologie Clinique et Expérimentale* **10** (1990) 147-156.
- LIPZSTEIN, J.L., BERTELLI, L., OLIVERIA, C.A.N., DANTAS, B.M., Studies of Cs retention in the human body related to body parameters and Prussian Blue administration, *Health Physics* **60** (1991) 57-61.
- MADSHUS, K., STRÖMME, A., Increased excretion of <sup>137</sup>Cs in humans by Prussian Blue, *Zeitschrift für Naturforschung A* **23** (1968) 391-392.
- MADSHUS, K., STRÖMME, A., BOHNE, F., NIGROVIC, V., Diminution of radiocaesium body-burden in dogs and human beings by Prussian Blue, *International Journal of Radiation Biology* **10** (1966) 519-520.
- MOESCHLIN, S., Thallium poisoning, *Clinical Toxicology* **17** (1980) 133-146.
- NIGROVIC, V., Enhancement of the excretion of radiocaesium in rats by ferric cyanoferrate (II), *International Journal of Radiation Biology* **7** (1963) 307-309.
- NIGROVIC, V., Retention of radiocaesium by the rat as influenced by Prussian Blue and other compounds, *Phys. Med. Biol.* **10** (1965) 81-91.
- NIGROVIC, V., BOHNE, F., MADSHUS, K., Dekorporation von Radionukliden (Untersuchungen an Radiocaesium), *Strahlentherapie* **130** (1966) 413-419.
- PAI, V., Acute thallium poisoning. Prussian Blue therapy in 9 cases, *West Indian Medical Journal* **36** (1987) 256-258.
- PEARCE, J., UNSWORTH, E.F., MCMURRAY, C.H., MOSS, B.W., LOGAN, E., RICE, D., HOVE, K., The effects of Prussian Blue provided by indwelling rumen boli on the tissue retention of dietary radiocaesium by sheep, *The Science of the Total Environment* **85** (1989) 349-355.
- PEARCE, J., HOVE, K., UNSWORTH, E.F., MOSS, B.W., A study of the use of indwelling boli containing Prussian Blue as a countermeasure to reduce the tissue radiocaesium content of sheep (in preparation).

- PEARCE, J., SMITH, J. Histopathological studies of the effects of indwelling rumen boli containing Prussian Blue in the sheep (in preparation).
- RAUWS, A.G., Thallium pharmacokinetics and its modification by Prussian Blue, *Naunyn-Schmiedeberg's Archives of Pharmacology* **284** (1974) 295-306.
- RICHARDS, J.I., HANCE, R.J., Countermeasures to the agricultural consequences of Chernobyl, *IAEA Bulletin* **42**, Vienna (1996).
- RICHARDS, J.I., HANCE, R.J., DARGIE, J.D., LUPIEN, J.R., WHITEHEAD, A.J., RANDELL, A.W., FIELD, C.B., "One decade after Chernobyl: the FAO response", *One Decade After Chernobyl - Summing Up the Consequences of the Accident (Proc. FAO/EC/IAEA Int. Conf. Vienna, 1996)*, Vienna (1996).
- RICHARDS, J.I., WEDEKIND, L., Reducing radiocaesium contamination of food products in the Chernobyl area, *IAEA Bulletin* **1**, Vienna (1993) 18-23.
- RICHMOND, C.R., BUNDE, D.E., Enhancement of caesium-137 excretion by rats maintained chronically on ferric ferrocyanide, *Proceedings of the Society of Experimental Biology and Medicine* **121** (1966) 664-670.
- ROBERTS, L., Radiation accident grips Goiania, *Science* **238** (1987) 1028-1031.
- STERNER, W., CHIBANGUZA, G., Akute orale Toxizität an Ratten mit Giese-Salz [ammonium (III) hexacyanoferrat (II)] (in Anlehnung an die OECD-Richtlinien), *IBR, Forschungs GmbH, Südkampen Nr.31,3030 Walsrode 1, Hannover (Projekt-Nr.:1-4-894-86)* (1987).
- TANG, M.H., GONG, Y.F., SHEN, C.Y., YE, C.Q., WU, D.C., Measurement of internal contamination with radioactive caesium released from the Chernobyl accident and enhanced elimination by Prussian Blue, *Journal of Radiological Protection* **8** (1988) 25-28.
- THOMPSON, D.F., Management of thallium poisoning, *Clinical Toxicology* **18** (1981) 979-990.
- VAN KESTEREN, R.G., RAUWS, A.G., DE GROOT, G., VAN HEYST, A.N.P., Thallium intoxication. An evaluation of therapy, *Intensiv. Med.* **17** (1980) 293-297.
- WENCKER, D., SPIESS, B., LAPP, C., LAUGEL, P., Influence of hexacyanoferrate (II) based treatments upon the elimination of heavy metal traces in urine. I. The case of lead, *Food Additives and Contaminants* **6** (1989) 351-357.
- WENCKER, D., SPIESS, B., LAUGEL, P., Influence of hexacyanoferrate (II) based treatments upon the elimination of heavy metal traces in urine. II. The case of cadmium, *Food Additives and Contaminants* **7** (1990) 375-379.
- WORLD HEALTH ORGANIZATION, "Anti-caking agents. Calcium, potassium, sodium ferrocyanide", *Toxicological Evaluation of Some Food Additives Including Anti-caking Agents, Antimicrobials, Antioxidants, Emulsifiers and Thickening Agents*, World Health Organisation, Geneva, *W1 W14H No. 5* (1974) 15-18.

## CONTRIBUTORS TO DRAFTING AND REVIEW

|  |   |
|--|---|
| Averin, V.S.                             | Belarus Institute of Agricultural Radiology, Gomel, Belarus   |
| Firsakova, S.K.                          | Belarus Institute of Agricultural Radiology, Gomel, Belarus   |
| Gurkov, V.V.                             | Department of Chernobyl Disaster Problems, Ministry of Agricultural Production, Minsk, Belarus  |
| Assimakopoulos, P.                       | Nuclear Physics Laboratory, University of Ioannina, Ioannina, Greece  |
| Hove, K.                                 | Department of Animal Science, Agricultural University of Norway, Ås-NLH, Norway   |
| Skuterud, L.                             | Norwegian Radiation Protection Authority, Østerås, Norway   |
| Strand, P.                               | Norwegian Radiation Protection Authority, Østerås, Norway   |
| Aleksakhin, R.M.                         | Radiation Biology and Medicine Section, Russian Institute of Agricultural Radiology and Agroecology, Obninsk, Kaluga Region, Russian Federation |
| Astasheva, N.                            | Institute of Extreme Situations in Agriculture, Moscow, Russian Federation  |
| Ratnikov, A.                             | Russian Institute of Agricultural Radiology and Agroecology, Obninsk, Kaluga Region, Russian Federation   |
| Travnikova, I.                           | Institute of Radiation Hygiene, St. Petersburg, Russian Federation  |
| Sobolev, A.                              | Ukrainian Institute of Agricultural Radiology, Kiev, Ukraine  |
| Prister, B.S.                            | Ukrainian Institute of Agricultural Radiology, Kiev, Ukraine  |
| Pronevich, V.                            | Sarni Research Station, Sarni, Rovno Region, Ukraine  |
| Pearce, J.                               | Food Science Department, The Queen's University of Belfast, Belfast, N. Ireland, United Kingdom   |
| Crick, M.                                | International Atomic Energy Agency  |
| Richards, J.I.<br>(Scientific Secretary) | FAO/IAEA Agriculture and Biotechnology Laboratory   |

**FAO/IAEA PUBLICATIONS ON AGRICULTURAL AND OTHER CONSEQUENCES OF THE  
CHERNOBYL ACCIDENT**

- [1] INTERNATIONAL ATOMIC ENERGY AGENCY, Principles for Establishing Intervention Levels for the Protection of the Public in the Event of a Nuclear Accident or Radiological Emergency, Safety Series No. 72, IAEA, Vienna (1985).
- [2] INTERNATIONAL ATOMIC ENERGY AGENCY, Derived Intervention Levels for Application in Controlling Radiation Doses to the Public in the Event of a Nuclear Accident or Radiological Emergency, Safety Series No. 81, IAEA, Vienna (1986).
- [3] FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS, Report of the Expert Consultation on Recommended Limits for Radionuclide Contamination of Foods, Rome, 1-5 December 1986, FAO, Rome (1987).
- [4] INTERNATIONAL ATOMIC ENERGY AGENCY, Revised Guidance on the Principles for Establishing Intervention Levels for the Protection of the Public in the Event of a Nuclear Accident or Radiological Emergency, IAEA-TECDOC-473, Vienna (1988).
- [5] INTERNATIONAL ATOMIC ENERGY AGENCY, Generic Intervention Levels for Protecting the Public in the Event of a Nuclear Accident or Radiological Emergency, Interim Report, IAEA-TECDOC-698, Vienna (1993).
- [6] INTERNATIONAL ATOMIC ENERGY AGENCY, Intervention Criteria in a Nuclear or Radiation Emergency, Safety Series No. 109, IAEA, Vienna (1994).
- [7] JOINT SECRETARIAT FOR THE BASIC SAFETY STANDARDS, International Basic Safety Standards for Protection against Ionizing Radiation and for the Safety of Radiation Sources, jointly sponsored by the Food and Agriculture Organization of the United Nations, the International Atomic Energy Agency, the International Labour Organisation, the Nuclear Energy Agency of the Organisation for Economic Co-operation and Development, the Pan American Health Organization and the World Health Organization, IAEA, Vienna (1994).
- [8] FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS/ WORLD HEALTH ORGANIZATION, Codex Alimentarius, General Requirements, Section 6.1, Guideline Levels for Radionuclides in Foods Following Accidental Nuclear Contamination, Joint FAO/WHO Food Standards Programme, Rome (1989).
- [9] INTERNATIONAL COMMISSION ON RADIOLOGICAL PROTECTION, 1990 Recommendations of the International Commission on Radiological Protection, Publication No. 60, Annals of the ICRP 21 1-3, Pergamon Press, Oxford and New York (1991).
- [10] INTERNATIONAL ADVISORY COMMITTEE, The International Chernobyl Project: Assessment of Radiological Consequences and Evaluation of Protective Measures, Technical Report by an International Advisory Committee, Section G, IAEA, Vienna (1991).
- [11] INTERNATIONAL ATOMIC ENERGY AGENCY/FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS, Guidelines for Agricultural Countermeasures Following an Accidental Release of Radionuclides, Technical Reports Series No. 363, IAEA, Vienna (1994).
- [12] INTERNATIONAL ATOMIC ENERGY AGENCY, Handbook of Parameter Values for the Prediction of Radionuclide Transfer in Temperate Environments, produced in collaboration with the International Union of Radioecologists, Technical Reports Series No. 364, IAEA, Vienna (1994).
- [13] WORLD HEALTH ORGANIZATION, EUROPEAN COMMUNITY, INTERNATIONAL ATOMIC ENERGY AGENCY, International Conference on 'One Decade After Chernobyl - Summing up the Consequences of the Accident', Vienna (1996).