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Bioaccumulation and decontamination mechanisms of persistent organic pollutants (PCB, DDT) in bodies of Bactrian camels

The present study aimed to determine the mechanisms of bioaccumulation and decontamination of Polychlorinated biphenyls (PCBs) and Dichlorodiphenyltrichloroethane (DDT) in the body of two-humped camels *Camelus bactrianus*. The experiment has been carried out in Suzak region of South Kazakhstan. Four lactating two humped camels received 0.8 mg of indicator PCBs (1.3 µg/kg body weight) and DDT 0.12 (DDT 0.2 µg/Kg body weight) mg per camel/day during two months and followed by a 4-month decontamination period. Milk and hump fat of experimental camels have been sampled. Milk samples were analyzed using a liquid-liquid and hump fat using solid extraction by gas chromatography and mass spectrometry method. Concentrations of PCBs and DDT in milk and hump reached a plateau at the end of the 2 months exposure period. Transfer rates into milk ranged between 2% for PCB 101 and 71 % for PCB 180 of the daily dose, which was generally lower than rates observed in ruminants. In the same time, the most important part of the contaminants has been stored in the humps. At the end of experimentation, the total quantity of PCBs excreted in milk was estimated to 28.6 µg and the total quantity accumulated during the contamination period in humps was 5530 µg. Despite a huge variability between the different congeners of iPCBs, the intermediate storage of lipophilic compounds in the humps reduced the concentrations excreted in milk but on the other hand would extend the duration of the decontamination period in comparison with ruminants.

Key words: bioaccumulation, decontamination, camels.

Introduction

Persistent organic pollutants (POPs) are organic compounds that are resistant to environmental degradation and capable of causing negative effects on human health and the environment [1]. Due to these properties, in 18 May of 2001, 110 countries signed the Stockholm Convention on Conference of United Nation Organization, where the countries agree: to prohibit and put out of production, use, and release of POPs. Initially, twelve POPs have been recognized as causing adverse effects on humans and the ecosystem and within are PCBs as industrial chemicals and DDT among pesticides [2]. There are 209 congeners of PCBs with different physical, chemical and biological properties. PCBs have been used in power and chemical plants; they have been included in

transformer and capacitor oils as additives to paints, plastics, rubber, as well as in lubricants and insulating materials. DDT is one of the pesticides, which included to the list of POPs. It is an effective insecticide widely used in agriculture over years during the last century to control the insect vectors of typhus, malaria and dengue fever. Also, it was made available to farmers as an agricultural insecticide [3]. The lactating ruminant may be exposed to DDT and PCB when they are eating contaminated feed or soil during grazing [4] as these compounds are accumulated in the body. According to the previous published data regarding impact of PCBs congeners 54, 80, 155 and one derivative of DDT 4,4 DDE in ruminant (sheep) previously contaminated by intramuscular injection under experimental conditions, the toxic equivalent of pollutants (on a fat basis) was approximately 2.5

times higher in milk than in blood [5]. Moreover, studies of the transfer of PCB to milk in goats exposed to a long-term intake of contaminated hay under experimental conditions also shown that the contaminants had rapidly high concentrations of PCBs in milk after one-week exposure [6]. These studies of kinetics of contamination and decontamination of the animals in order to precise the transfer of pollutants in lactation goats and sheep were carried out in European countries. But researches on the transfer of pollutants and the mechanism of distribution of contaminants in camel organs (hump-fat, milk) have never been carried out and the concentration of this pollutant has not been studied in comestible parts of animals. Camels have a special characteristic as a biological model among all farm animals, and in general all mammals. Camels have the ability to survive and adapt to hard environmental conditions. Metabolic studies of PCBs and DDT in the body of *Camelus bactrianus* allow to understand the adaptive ability to survive in polluted environments. In the comparative studies of the effect of organic and inorganic selenium supplementation on selenium status in camel, metabolism of selenium in camel organism is observed to be less than in cattle [7]. Physiological characteristics of laboratory animals are considered from the standpoint of comparison with human physiology. Impact of these pollutants helped get a general idea, how they can affect the humans. Studies on the sheep and goats were conducted to control the meat of these animals as the object of the food chain. On the one hand, these studies supplement scientific data as a potential contamination object in the food chain. On the other hand, studies on such special biological models as *Camelus bactrianus* allow to better understand the biological intake of pollutants such as PCBs and DDT. In addition, it is necessary to take into account the fact that in the desert regions camels are sometimes the only type of livestock; as a result they are the only source of milk, meat and wool for humans. That's why this work devoted to study the entry and distribution of DDT and PCBs in the body of camels, as well as ways of removing these contaminants.

Material and methods

The main three steps of this experiment were: (1) contamination of the animals to reach a steady state situation, (2) determination of the POPs concentration in the different compartments (blood, fat and milk), and (3) monitoring of the decontamination process. Regarding the first step, it was necessary to

assess the importance of the different compartments: (1.1) weight of the animal, (1.2) weight of the hump as main site of fat storage, (1.3) milk production (especially its fat content). Regarding the second and the third steps, the changes of the POPs concentrations during contamination and decontamination stage are assessed in the different compartments.

For experiment four lactating *Camelus Bactrianus*, 7-16 years old were used. The weight of animals ranged from 400 to 520 kg. Before experiment, data about age, calving date, and parity were reported as well as sex of calves. All camels have been identified with ear tags. The animals were in healthy conditions all along the study.

Experimental camels were exposed to DDT (Pestanal, analytical standard – 31041, Fluka) and PCBs mixture (Aroclor 1254 – analytical standard- 48586, Sigma-Aldrich), which were introduced in gelatin capsules (length – 2 cm. diameter – 9 mm) by hexane solvent. The capsules were filled with icing sugar fixing the introduced chemicals and allowing the evaporation of the solvent. The contaminants for one camel were quantified for PCBs 1.3 µg/kg and DDT 0.2 µg/kg body weight by day. In one capsule the concentration of PCBs was 0.8 mg and DDT 0.12 mg per camel/day. As each camel received one capsule during 56 days, the total exposure doses of one camel was 44 mg of PCBs and 6.7 mg of DDT. The daily supply of capsules was realized inside of bread. In order to reach the concentration plateau (steady state) more rapidly, a primary dose of 9.13 mg for PCBs and 1.41 mg DDT was given by intravenous injection on the first day of exposure. This dose with PCBs and DDT solution was prepared in oil solvent (Cremophor EL - reference 95921 SUPELCO). The primary dose was 12 times higher than dose in capsules. During the experiment the milk, serum of blood and hump fat were sampled. Also, the body, hump measurements were made and milk yield was estimated, the milk composition (fat content, dry matter and density) was determined at each sampling date.

Analytical works have been done in CPHMA (The Center of Physico-Chemical methods of analysis), Laboratory of Ecology of the Biosphere, in GH-Agilent, with mass spectrometric and flame ionization detection Agilent 6890N / 5973N, equipped with a system of pre-concentration of liquid and solid samples Agilent-Velocity XPT.

Milk and blood serum were analyzed using a liquid-liquid and fat using solid extraction followed by cleanup on a multi-layer silica gel column, evaporative concentration to 20 µL and analysis on 7890A/5975C TAD TVL GC-MS (Agilent, USA)

equipped with Combi-PAL autosampler (CTC Analytics AG, Switzerland). Two μL of sample was injected to split/splitless inlet heated to 250°C in splitless mode. Separation was done on a DB-5MS 60 m x 0.25 mm, 0.25 μm film column (Agilent, USA) at a constant flow of helium (purity 99.995%, Orenburg-Tehgas, Russia) equal to 1 mL/min. Detection was done in selected ion monitoring mode (SIM) using 6-group program for detection of target ions. PCB209 was used as internal standard spiked to samples in amount of 300 pg.

The results have been expressed by the mean of four camels within three periods \pm standard error of the mean (SEM): period 1 – contamination period; period 2 – first two months of decontamination period with fat mobilization; period -3 – second two months of decontamination period with fat storage. The statistical differences between the 3 periods were assessed by variance analysis (ANOVA) using XLstat software (Addinsoft ©). Only the difference between periods was tested.

Results and their discussion

The metabolism of POPs includes the intake, the transport of biological fluid in blood and lymph, their storage in adipose tissue and the excretion through feces, urine and milk. In the frame of our experiment the concentration in hump fat and excretion in milk has been assessed in order to determine the bioaccumulation and decontamination mechanisms of pollutants in these different compartments. In the gastrointestinal tract, after ingestion of the capsule with contaminants, pollutants enter into forestomach of the camel, and then entered in the bloodstream. The blood transferred the pollutants to other compartments, especially in adipose tissue, the hump representing the main part. A part of the contaminants is excreted with milk in lactating ruminants and probably also through the feces. For a better understanding, the results were expressed according to the 3 main periods of the experiment: (1) the mean values during the two months of contamination (contamination period), (2) the mean values during the first 2 months of decontamination, and (3), the mean values during the last two months of decontamination. However, the kinetics was presented by taking into account the mean of the 4 camels and the sum of PCBs on the one hand and of DDT/DDE on the other hand.

At the beginning of the contamination period, the lipophilic properties of pollutants lead to a rapid in-

creasing of their concentrations in hump, and because the animals are in phase of fat storage, in total quantity. At the same time, the concentrations in milk did not increase in a notable manner. When the plateau was reached after two months of contamination, the concentrations in blood and milk increased, showing the elimination of pollutants (Fig. 1, 2).

It seems that the main storage of organic pollutants in the hump would first slow the transfer into milk but also extend the time necessary for decontamination in comparison to other ruminants.

By considering the cumulative excretion in milk all along the experiment and the quantity of pollutants in hump at the beginning of the experiment, the global kinetics of bioaccumulation and excretion process could be summarized for both PCB and DDT (Fig. 1,2).

This phenomenon is accentuated because of the hump weight decreased after starting decontamination (during summer time) due to the fat mobilization. The concentration and the quantity of pollutants stored in hump decreased regularly all along the decontamination period. The elimination in milk appeared low in quantity because the transfer to milk is in low percentage (between 2 and 9% depending to congeners) contrary to other species as cow and goat. A similar trend occurred for PCBs and DDT.

At the end of experimentation, the total quantity of PCB and DDT excreted in milk were estimated to 28.6 and 0.95 μg respectively and the total quantity accumulated during the contamination period in hump was 5530.4 and 54.3 μg respectively. In consequence, the percentage of excreted pollutants in milk was low: only 0.52% for PCB and 1.74% for DDT on average. The percentage of pollutants accumulated in hump was less than 15% of the total intake with a higher proportion for PCB than for DDT. After 4 months of decontamination, the total quantity of PCB and DDT was disappearing respectively 47.4% and 35.5% of the maximum concentration at the contamination period.

Conclusion

Besides the assessment of the live weight, hump volume and milk yield in field conditions, the main conclusions of our work regarding the transfer of POPs in Bactrian camel model are as follows:

The role of the camel hump (from 5.3 to 21.5 kg) as a pivotal organ (due to its importance in the cycle lipid storage/lipid mobilization) in the metabolism of pollutants having lipophilic properties is verified.

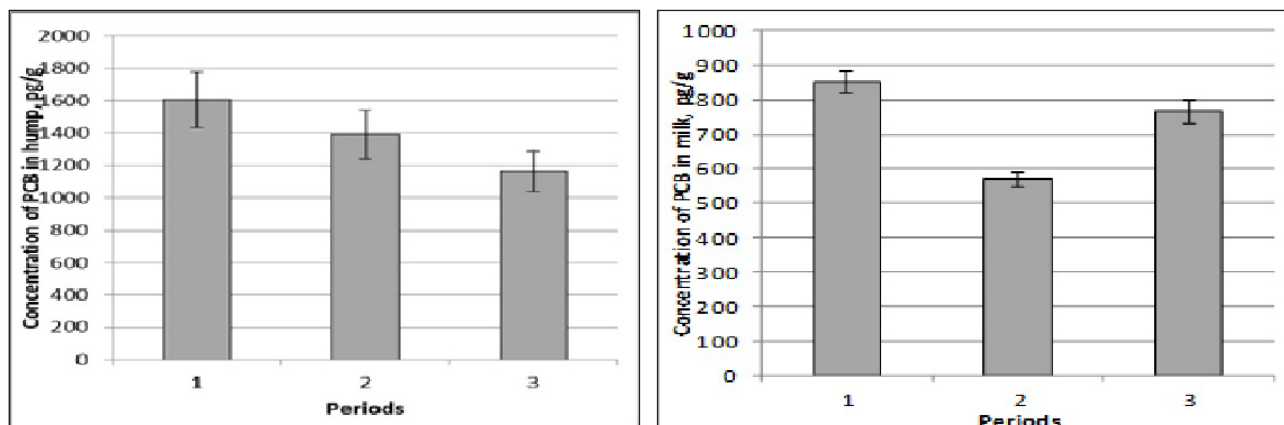


Figure 1 – Mean concentration of PCBs and SEM in hump tissue (left side) and in milk (right side) during the 3 periods of experimentation: 1 – contamination; 2 – first two months of decontamination; 3 – last two months of decontamination

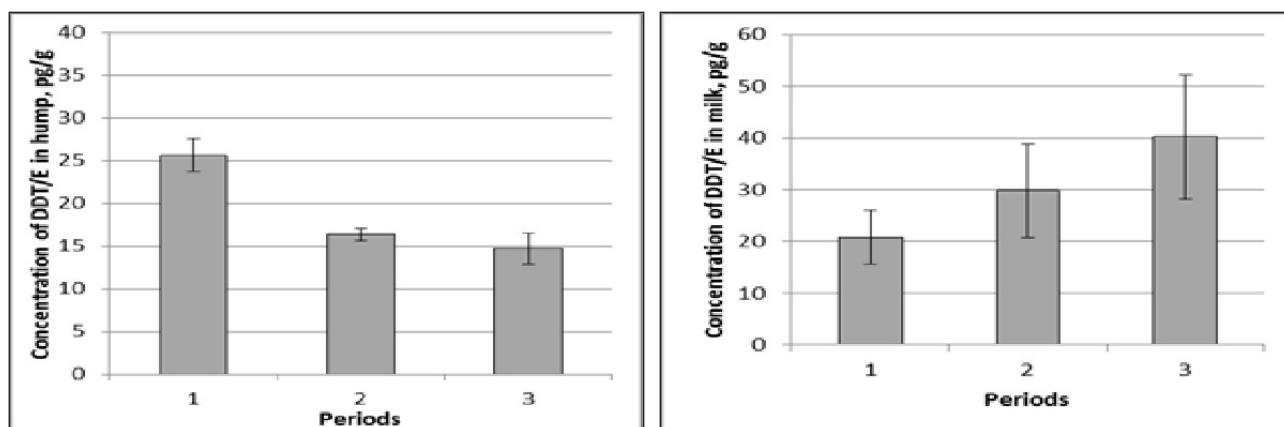


Figure 2 – Mean concentration of DDT/E and SEM in hump tissue (left side) and in milk (right side) during the 3 periods of experimentation: 1 – contamination; 2 – first two months of decontamination; 3 – last two months of decontamination

At reverse, in spite of the importance of this route of excretion and thanks to its fat content, only small concentrations of pollutants are observed in milk. On average, after 6 months of experiment, the percentage excreted in milk was 0.52% (PCBs) and 1.74% (DDT) of the cumulative POPs in the hump, but there is a high variability between congeners.

Based on the maximum quantity of pollutants in hump during the contamination period and the quantity available at the end of experiment, the percentage of loss of PCB was 47.4% and for DDT, it was 35.5% that means the camel could be completely decontaminated within less than one year.

Moreover, based on literature data, the concentrations of pollutants in milk were low compared to milk from other contaminated dairy animals as goats and cows. The carry-over rate (COR) was 8.9% for PCB52 in our study vs according to the literature 25% in goats, and 7.7% for PCB180 in our study vs

55% in goats and 65% in cows. As the carry over rate for camels seems to be very low, in comparison to other ruminants, we could conclude that:

- The camels would transfer pollutants in milk slower than other ruminants;
- The application of transfer rates stated in other ruminants may overestimate the exposure of dairy camels.
- Complete decontamination of camels would certainly take more time than in other ruminants

The present work has been achieved in a private farm. The lack of experimental camel farms in Research structures of Kazakhstan is an important constraint for the future research activities regarding this specie. Regarding the important place of Camel products in Food habits in Kazakhstan, this lack should be considered in future development of research facilities. In the international scientific community interested by the camel (International Society for Camelid

Research and Development –ISOCARD), the studies regarding the behavior of camel faced to pollution are very few. The present study appears original and innovative for camel scientists over the world and confirms the interest of this species as a biological model in such research regarding the impact of environmental pollution on animal products. The special focus on Bactrian Camels poorly studied in the scientific literature allowed to enlarge existing knowledge about this specie emblematic of Central Asia.

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