A common viral infection may change target organ distribution of 2,3,7,8-TCDD

Funseth, E.[A], Ilbäck, N-G.[B]
[A]Toxicology Division, National Food Administration
[B]Department of Infectious Diseases, Uppsala University, University Hospital, Uppsala, Sweden

Introduction

Immune mechanisms have in several studies been shown to be vulnerable to the effects of 2,3,7,8-TCDD. However, this is a complex immunomodulating chemical, and although the effects have been studied on different immunological parameters[1] the exact mechanisms are not yet fully explained. Results from several studies indicate that TCDD and related compounds may impair host resistance to both viral and bacterial pathogens.[2-6] Changes in host-parasite interactions may also include changed uptake and distribution of environmental pollutants.[7,8]

Coxsackievirus type B3 (CB3) belongs to a group of viruses that is common in our environment, and virtually all humans become infected with these viruses during their lifetime.[9] Coxsackievirus B3 may sometimes cause myocarditis, and possibly also type I diabetes in humans.[9] The murine model of CB3 has a well-described pathogenesis that closely mimics this disease in humans.[9,10]

We have previously shown that challenge with this virus (CB3) will lead to changed tissue uptake and distribution of environmental pollutants, such as nickel and cadmium.[7,8] An increased tissue uptake and distribution may subsequently also increase target organ toxicity and further decrease resistance to this invading micro-organism. The present study was performed to investigate whether this common human viral infection (CB3), here adapted to the mouse, affects tissue uptake and distribution of 2,3,7,8-TCDD.

Materials and methods

Mice: Male A/J-mice were bred and maintained at the Division of Toxicology, National Food Administration, Uppsala, Sweden. The mice were housed six per cage at 23±1°C on a 12-hour light/dark cycle behind strict hygienic barriers with free access to food (R3; Ewos, Södertälje, Sweden) and water. The mice were randomized into groups of similar initial mean body weight. Infected and control mice were studied simultaneously.

Virus: A stock solution of CB3 virus was diluted with phosphate buffered saline (PBS) and each mouse was inoculated intraperitoneally with $2x10^4$ infectious particles in 0.2 ml PBS.

Test substance: Radioactively labelled ^{14}C-TCDD (2,3,7,8-tetrachlorodibenzo-*p*-dioxin; specific activity 122 mCi/mmol) was purchased from Cambridge Isotope Laboratories, Cambridge, MA, USA. The substance was dissolved in a mixture of toluene/DMSO (v/v = 1/10) to give 0.5 μCi/10 μl.

Treatment and tissue sample collection: The mice were inoculated with virus on day 0. On days 4 and 7 *post* inoculation (*p.* inoc.), infected mice and corresponding uninfected control mice were injected intravenously in a tail vein with 0.5 μCi of ^{14}C-TCDD, corresponding to approximately 50 μg TCDD/kg. The mice were sacrificed 24 hours after the ^{14}C-TCDD administration, and the spleen, thymus, heart, pancreas and liver were dissected, weighed and counted for radioactivity in the entire organs.

Liquid scintillation counting: The TCDD-content of the tissues was determined by means of standard liquid scintillation techniques. In brief, tissue samples were cut into pieces and digested in Biolute S at 50°C for four hours. Samples that contained blood were bleached with isopropanol and 35% H_2O_2 at 50°C for two hours. Finally, scintillation fluid was added, and the samples were counted for radioactivity in a liquid scintillation counter.

Statistical analysis: The effects of infection on ^{14}C-TCDD-accumulation in selected tissues were calculated using Student´s t-test which compared the uninfected control group with the infected group.

Results

The uptake of TCDD in the spleen increased significantly during this viral infection (Table 1), and was most pronounced on day 4 after the inoculation, i.e. during the acute phase of the disease when viraemia peaked. In the thymus, no difference in TCDD-uptake was observed between infected and control mice. The concentration of TCDD was however increased, and by day 7 it was 50% ($p<0.001$) higher than in the control mice (data not shown).

The pancreas showed an early TCDD-uptake on day 4, whereas the uptake later during the disease, i.e. on day 7, was comparable to that in the uninfected control group (Table 1).

Table 1. The uptake of ^{14}C-TCDD when expressed as cpm/entire organ (n=8 for controls and n=9 for mice in the infected group). Values are given as mean±SE. Statistically significant differences ($^{*}p<0.05$; $^{**}p<0.01$; $^{***}p<0.001$) between uninfected control and infected groups are denoted by asterisks.

	Spleen	Thymus	Pancreas	Heart	Liver
Controls	131±39	108±36	248±120	88±10	30900±9400
Day 4 *p.* inoc.	1030±920**	124±46	461±230*	193±83**	85000±52000*
Day 7 *p.* inoc.	209±31***	90±25	251±110	122±18***	39500±10500

Also the heart showed a peak of TCDD uptake on day 4, but there was still a considerable uptake on day 7, when inflammatory lesions develop. Similarly, the uptake in the liver was most pronounced on day 4 (Table 1).

Discussion

The results indicate that there is an increased TCDD-uptake in the selected tissues during this common viral infection (CB3), and that this increased tissue uptake seems to be most pronounced during the viraemic phase of the disease, i.e. day 4 *post* inoculation.

It also seems reasonable to assume that challenge with pathogenic micro-organisms will cause changed tissue uptake and distribution of environmental pollutants.[7,8] Several infections, including CB3, may cause lipid accumulation in organs.[10] Considering the lipid solubility of TCDD, a potential risk exists of further TCDD-accumulation in lesions during inflammatory disease and a subsequently increased toxicity in vital organs. The TCDD-uptake in the target organs of this CB3 virus infection, i.e. the heart and pancreas, seemed to peak on day 4 of the infection, corresponding to the peak of viraemia. Similarly, the spleen showed the highest uptake at this point of the disease. It is thus possible that this early uptake of TCDD has an immunomodulating effect that affects progression of the disease.[11,12]

The total TCDD-uptake in the thymus was not changed during this infection, while the concentration in the organ was significantly increased. This is probably due to the normally occurring emptying of thymocytes into the blood during generalized infection. The effects of this increased TCDD-concentration on future function and maturation of T-cells is, however, not clear.

The impact of the changed tissue uptake and distribution of TCDD during this common viral (CB3) infection may have a significant effect on future organ function. It is also possible that this will subsequently increase target organ toxicity and further decrease resistance to invading microorganisms.

References

1 Vos JG, Luster MI. Immune alterations. In: R.D. Kimbrough RD, Jensen AA, eds. *Halogenated biphenyls, terphenyls, naphtalenes, dibenzodioxins and related products.* Amsterdam, Elsevier Science Publishers B.V. 1989:295-322.

2 House RV, Lauer LD, Murray MJ. Examination of immune parameters and host resistance mechanisms in B6C3F1 mice following adult exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin. *J Toxicol Environ Health* 1990;31:203-15.

3 Imanishi J, Haruko N, Motoo M *et al.* Effect of polychlorinated biphenyl on viral infections in mice. *Infection and immunity* 1980;29:275-7.

4 Luster MI, Boorman GA, Dean JH *et al.* Examination of bone marrow, immunologic parameters and host susceptibility following pre- and postnatal exposure to 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD). *Int J Immunopharmac* 1980;2:301-10.

5 Thomas PT, Hinsdill RD. Effect of polychlorinated biphenyls on the immune responses of Rhesus monkeys and mice. *Toxicol Appl Pharmacol* 1978;44:41-51.

6 Vos JG, Kreeftenberg JG, Engel HW *et al.* Studies on 2,3,7,8-tetrachlorodibenzo-*p*-dioxin-induced immune suppression aand decreased resistance to infection: endotoxin hypersensitivity, serum zink concentrations and effect of thymosin treatment. *Toxicology* 1978;9:75-86.

7 Ilbäck N-G, Fohlman J, Friman G. A common viral infection can change nickel target organ distribution. *Toxicol Appl Pharmacol* 1992;113:00-00.

8 Ilbäck N-G, Fohlman J, Friman G, Wicklund-Glynn A. Altered distribution of ^{109}cadmium in mice during viral infection. *Toxicology* 1992;71:193-202.

9 Woodruff JF. Viral myocarditis. A review. *Am J Pathol* 1980;101:425-83.

10 Ilbäck N-G, Mohammed A, Fohlman J, Friman G. Cardiovascular lipid accumulation with Coxsackie B virus infection in mice. *Am J Pathol* 1990;136:159-67.

11 Funseth E, Ilbäck N-G. Effects of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin on blood and spleen natural killer (NK) cell activity in the mouse. *Toxicol Letters* 1992;56:00-00.

12 Lundberg K, Grönvik K-J, Goldschmidt TJ *et al.* 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) alters intrathymic T-cell development in mice. *Chem Biol Interactions* 1990;74:179-93.