CADMIUM ACCUMULATION AND METALLOTHIONEIN-LIKE BINDING ACTIVITY IN CARDIAC AND SKELETAL MUSCLE OF *SQUALUS ACANTHIAS*

Marlies Betka and Gloria V. Callard
Department of Biology, Boston University, Boston MA 02215

Although renal and hepatic mechanisms of cadmium (Cd) toxicity have been extensively studied, there is good evidence that Cd may target the heart directly. For example, epidemiological studies have linked high environmental Cd levels to high mortality from heart disease (Carrol, JAMA 198:177-183, 1966) and to increased incidence of deaths from hypertensive disease, ischemic heart disease and stroke (Neuberger et al., Sci. Tot. Envir. 94: 261-272, 1990). As part of a program of research in which we are using the shark *Squalus acanthias* to study effects of Cd on male reproduction, we observed that cardiac muscle accumulated a tracer dose of ^{109}Cd to a much greater extent than skeletal muscle (9,300 vs. 513 cpm/g, 24 h postinjection), but levels were in the same range as those in liver (14,277cpm/g), a recognized Cd target. Additionally, ^{109}Cd concentrations in heart were retained virtually unchanged up to 7 d after tracer injection, despite the disappearance of ^{109}Cd from the peripheral plasma (125 cpm/ml). These data indicate a long biological half-life for Cd in heart and the possibility that *S. acanthias* may have utility as a model for studying low-dose, cumulative effects on cardiac physiology. Here we report attempts to further characterize the Cd-accumulating mechanism of *S. acanthias* heart and describe the presence and Cd-inducibility of a metallothionein (MT)-like ^{109}Cd-binding component.

Sharks were left untreated or injected with a single dose of radioinert $CdCl_2$ (5 mg/kg, i.v.) 3 d before killing (3/treatment group). Ventricular and skeletal muscle was stored at -70°C until analysis. To measure actual Cd burden, tissues (200 mg) were digested with nitric acid and Cd measured using a Perkin-Elmer 5100 PC atomic absorption spectrophotometer. MT-like ^{109}Cd-binding activity was measured in heat-treated cytosolic subfractions using a standard Cd/hemoglobin affinity assay (Eaton & Toal, Toxicol. Appl. Pharmacol. 66:134-142, 1982). Statistical analysis was performed using Student's t-test.

In untreated controls, the Cd burden of cardiac and skeletal muscle did not differ significantly (0.14 ± 0.10 vs. 0.12 ± 0.12 μg/g, respectively), and differences in the quantity of MT-like proteins in the two tissues were marginal (0.23 ± 0.04 vs. 0.10 ± 0.02 μg/g, respectively, $p < 0.05$). By contrast, a single injection of Cd 3 d before killing increased Cd levels in cardiac muscle 72-fold (to 10.43 ± 0.54 μg/g, $p < 0.001$ vs. controls) but only 3-fold in skeletal muscle (to 0.35 ± 0.09 μg/g, not significant). Moreover, the cytosolic MT-like binding capacity of cardiac muscle was elevated 23-fold after Cd-pretreatment (to 5.20 ± 0.49 μg/g, $p < 0.001$ vs. controls), whereas MT-like components of skeletal muscle were unchanged by prior Cd exposure. We conclude that cardiac muscle of *S. acanthias* is a direct target of Cd action, as measured by inducibility of Cd binding activity and consequent increased accumulation/retention of Cd. Further studies are required to determine whether this biological response is the first step in an effector pathway leading to perturbations of cardiac function. This work was supported by a MDIBL Young Investigator Fellowship (MB) and NIEHS P42 ES-07381 (GVC).