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Figure SI-1. Hepatic distribution profiles of eight selected metals (a – Co, b – Cu, c – Fe, d – Mn, e – Mo, f – Se, g – Zn, and h – Cd) among cytosolic proteins of different molecular masses in Vardar chub (*Squalius vardarensis*) caught in two Macedonian rivers (Bregalnica and Zletovska). The profiles were obtained by separation of hepatic cytosols by SEC₂₀₀-HPLC. The results are presented as nanograms of metals eluted at one minute intervals. Four fish were used for these analyses (No. 1, 2, 3, and 4), and their total cytosolic metal concentrations in Vardar chub liver are presented within the figure. The MMM-peaks containing predominantly Fe (t_e 26-29 min) and the LMM-peaks containing predominantly Cd, Cu and Zn (t_e 30-34 min), which are marked by dotted lines, were further collected for fish No. 1-3 and used for AEC-HPLC analyses.



Figure SI-2. Gill distribution profiles of four selected metals (a – Fe, b – Se, c – Zn, and d – Cd) among cytosolic proteins of different molecular masses in Vardar chub (*Squalius vardarensis*) caught in two Macedonian rivers (Bregalnica and Zletovska). The methodology, the analyzed samples and the result presentation are the same as in the Figure SI-1. The MMM-peaks containing predominantly Fe (t_e 26-29 min) and the LMM-peaks containing predominantly Cd and Zn (t_e 30-34 min), which are marked by dotted lines, were further collected for fish No. 1-3 and used for AEC-HPLC analyses.



Figure SI-3. SEC₂₀₀-HPLC chromatograph of standard protein transferrin (human, concentration 2 mg mL⁻¹) recorded by UV detection at 280 nm.



Figure SI-4. Mass spectra obtained by MALDI-TOF-MS for MMM Fe-binding biomolecules, which were separated by SEC_{200} -HPLC from hepatic (a) and gill (b) cytosols of Vardar chub, as well as for standard proteins transferrin (c) and superoxide dismutase (d).



Figure SI-5. Hepatic distribution profiles of Cd (a, d), Zn (b, e) and Cu (c, f) and gill distribution profiles of Cd (g, i) and Zn (h, j) among cytosolic proteins and heat stable cytosolic proteins of different molecular masses in Vardar chub (*Squalius vardarensis*) caught in two Macedonian rivers (Bregalnica and Zletovska). The profiles were obtained by separation of hepatic and gill cytosols and heat-treated cytosols by SEC₇₅-HPLC. Two fish were used for these analyses (No. 2 and 5) and the results are presented as nanograms of metals eluted at one minute intervals. The LMM-peaks of heat-treated hepatic cytosols (L-Cd-peaks), which contained Cd, Cu and Zn (t_e 21-25 min) and were marked by dotted lines, were further used for AEC-HPLC analyses. The LMM-peaks of heat-treated gill cytosols (G-Cd-peaks), which contained Cd (t_e 21-25 min) and were marked by dotted lines, were further used for direct MS analyses.



Figure SI-6. Hepatic distribution profiles of Mo (a, c) and Se (b, d) and gill distribution profiles of Mo (e, g) and Se (f, h) among cytosolic proteins and heat stable cytosolic proteins of different molecular masses in Vardar chub (*Squalius vardarensis*) caught in two Macedonian rivers (Bregalnica and Zletovska). The methodology, the analyzed samples and the result presentation are the same as in the Figure SI-5. The VLMM-peaks of heat-treated hepatic and gill cytosols, which contained predominantly Mo and traces of Se (t_e 26-30 min) and which were marked by dotted lines, were further used for direct MS analyses.

