DMAC-Analysis

The photometric analysis was done according to Prior et al. (2010). In short: the samples were dissolved or diluted in acetone+water+acetic acid (74.5+25+0.5, v+v+v) and incubated with the coloring agent (4-dimethylaminocinnamaldehyde (DMAC)) in acidified ethanol. The absorbance was read at 545 nm. For quantifying a calibration curve of procyanidin A2 was used.

HPLC-Analysis

For analysis of the procyanidins by UHPLC, the juice was filtered and analyzed without further treatment. The granulate was dissolved in acteone+water (70+30, v+v). For analysis of the procyanidins an Acquity UPLC system by Waters (Milford, MA, USA) consisting of a binary pump (BSM), an autosampler (SM; cooled to 10 °C), a column oven (CM) set at 40 °C, a diode array detector (PDA), and a triplequadrupole mass spectrometer (Acquity TQD) with electrospray interface operating in negative mode was used. An Acquity BEH Shield RP18 column (150 mm × 2.1 mm, 1.7 μ m; Waters) was used for separation. The whole system was controlled by MassLynx 4.1 software. The solvents were LC-MS grade water with 0.1% (v/v) formic acid (mobile phase A) and acetonitrile with 0.1% (v/v) formic acid (mobile phase B). The UHPLC gradient was as follows:

0–28 min, 98–76% A; 28– 29 min, 76–0% A; 29–31 min, 0% A; 31–33 min, 0–98% A; 33–35 min, 98% A; flow rate = 0.4 mL/min.

Two microliters of each sample extract was injected. For quantification of A-type procyanidins, the mass spectrometer was tuned using a standard solution of procyanidin A2. The resulting parameters were as follows: capillary voltage, –2.0 kV; cone voltage, 46 V; extractor voltage, 2.0 V; RF voltage, 0.20 V; source temperature, 150 °C; desolvation temperature, 450 °C; cone gas (nitrogen) flow, 50 L/h; desolvation gas (nitrogen) flow, 800 L/h.

For quantification purposes mass traces of procyanidins m/z were measured by using selected reaction monitoring (SRM) with the following compound-specific transitions of parent and product ions: A-type dimers m/z 575 \rightarrow 449, A-type trimers m/z 863 \rightarrow 575 and 863 \rightarrow 573. The dimmers and trimers were quantified as procyanidin A2 dimer equivalents (A2 equiv) with an external calibration curve of procyanidin A2 in the range of 10–100 µg/mL.

All UHPLC-MS² analyses of each sample were done in duplicate.

Mass traces of A-type dimers (m/z 575 -> 449) and A-type trimers (m/z 863 -> 575 and 863-> 573) in lingonberry granulate and lingonberry juice. Peak labels according to Jungfer et al. 2012.