Food preservatives characterized by an orthogonal effect-directed analysis system that hyphenates NP-HPTLC, bioassay, RP-HPLC and ESI-MS

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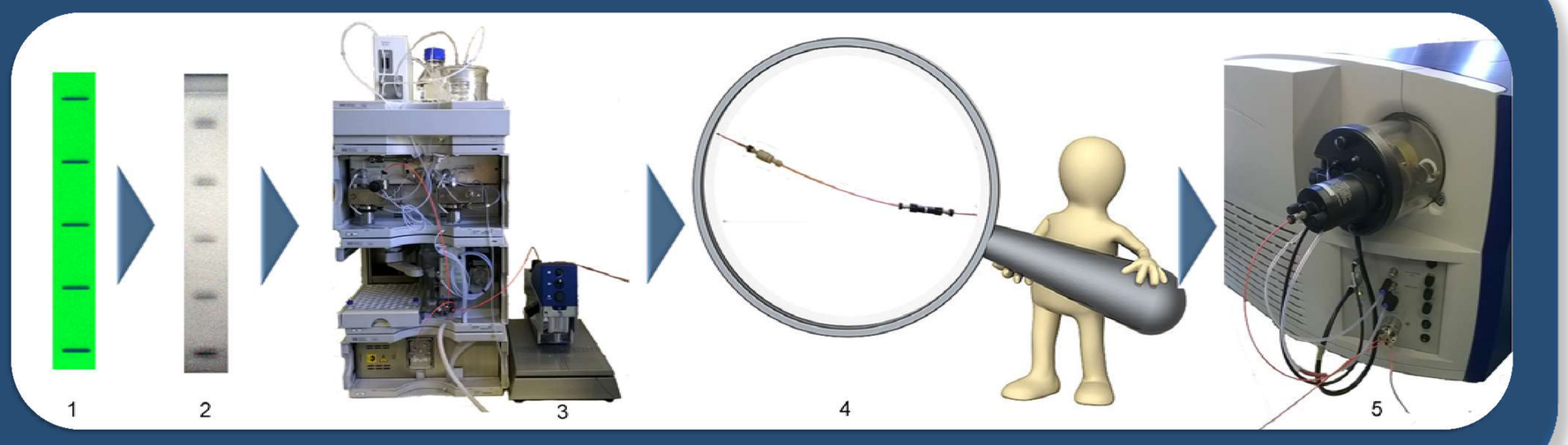
Highlights

- Only bioactive zones of interest are analyzed, which provides high specificity and selectivity and streamlines the analytical workflow.
- The effect is directly linked to the bioactive compound that was detected through HPTLC in combination with bioassays (effect-directed analysis, EDA).
- Many samples can be analyzed in parallel under identical analytical conditions → effective bioprofiling of food, supplements and cosmetics
- This hyphenated method is able to cope with coeluting substances.

Goal From non-target bioprofiling to substance identification in a single sample run!

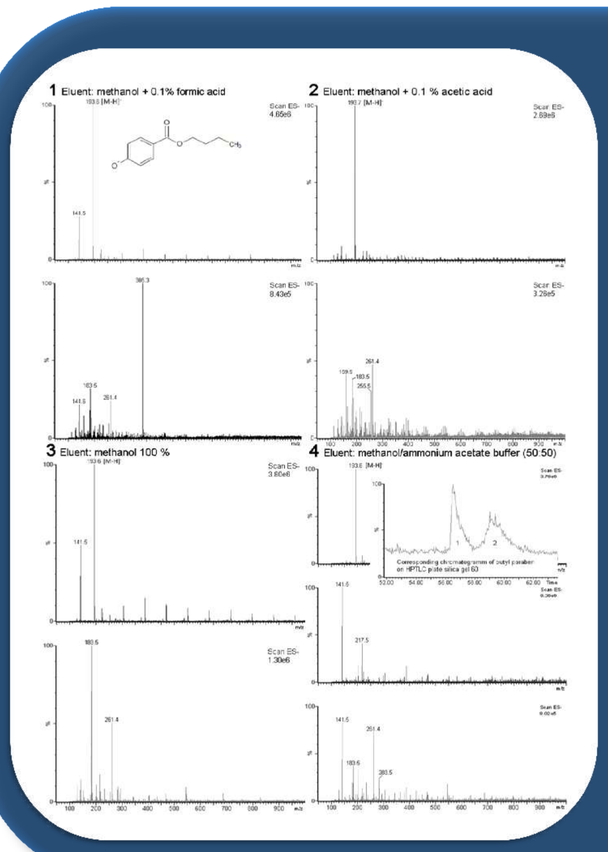
Problem High salt load of the bacterial suspension adsorbed on the HPTLC plate interferes with MS detection

Workflow: After application of the preservatives on a HPTLC plate (1) and detection via *Aliivibrio fischeri* (2), zones were eluted with the TLC-MS Interface (CAMAG, 3) and transferred over a monolithic RP-18 column (Merck, 4) and a Rheodyne valve (for deposition of the salt load into the waste) into the ESI-MS (Waters Quattro Ultima, 5).



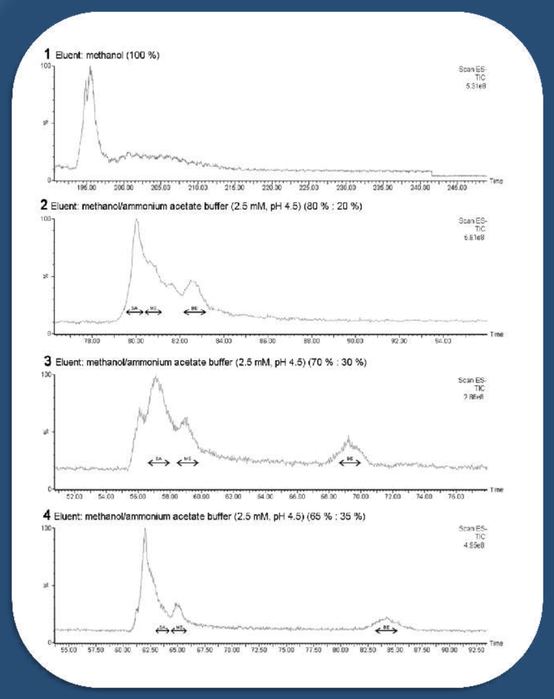
Method development

Exemplarily demonstrated for analysis of food preservatives detected by *Aliivibrio fischeri*



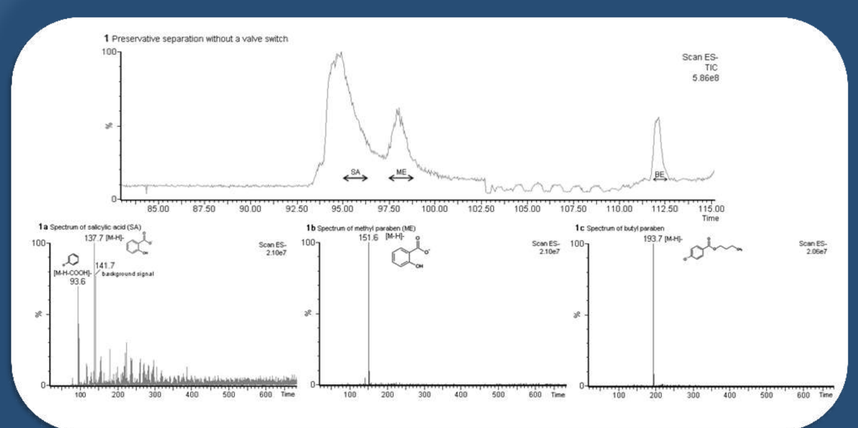
1st step: Eluent selection

Four eluents were studied for ionization, exemplarily of butyl paraben (*m/z* 193, 1-4). The eluents were compared with regard to their resulting signal intensities and background signals. The best result was achieved with a mixture of methanol and 2.5 mM ammonium acetate buffer, 70:30. This eluent was used for further measurements.



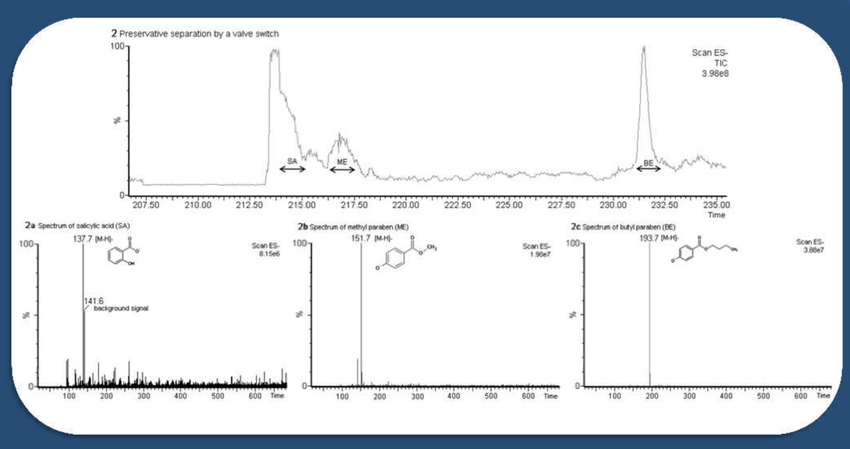
2nd step: Separation

A preservative mixture, *i. e.* salicylic acid (SA), methyl paraben (ME) and butyl paraben (BE) was applied on a NP-HPTLC plate, eluted and separated via a RP-18 column. The gradient was optimized starting with methanol (1), followed by three mixtures with 2.5 mM ammonium acetate buffer 80:20 (2), 70:30 (3) and 65:35 (4).



3rd step: Splitting of the salt load

By turning the switching valve into operation mode the salt load was deposited for a defined time period into the waste. In doing so, the TIC of the initial separation (1) of 5.86 E8 was reduced to 3.98 E8 (2). Also the analytes showed a higher intensity. Quod errat demonstrandum!



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