

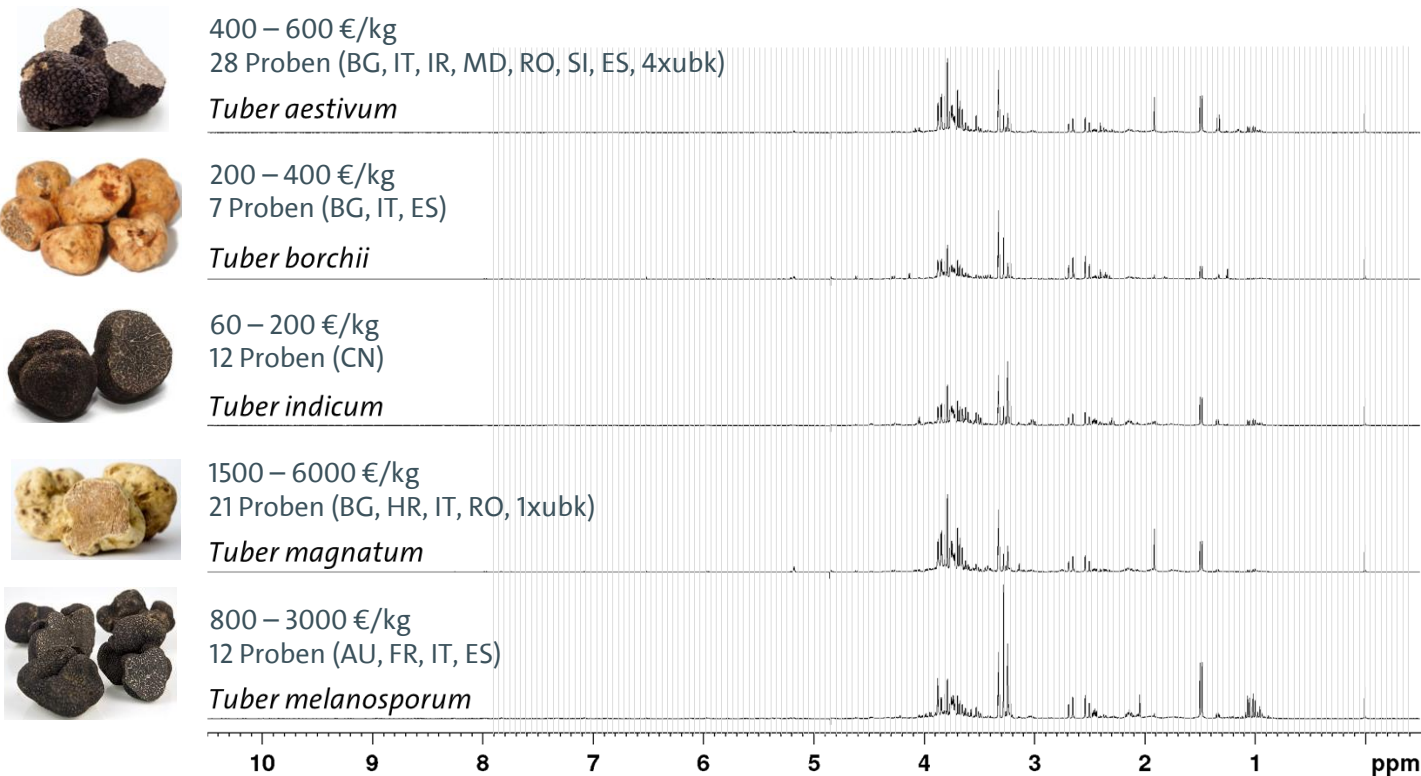
Thomas Hackl

Praktische Probleme der Kernresonanzspektroskopie 2023 | FMP Buch | Berlin

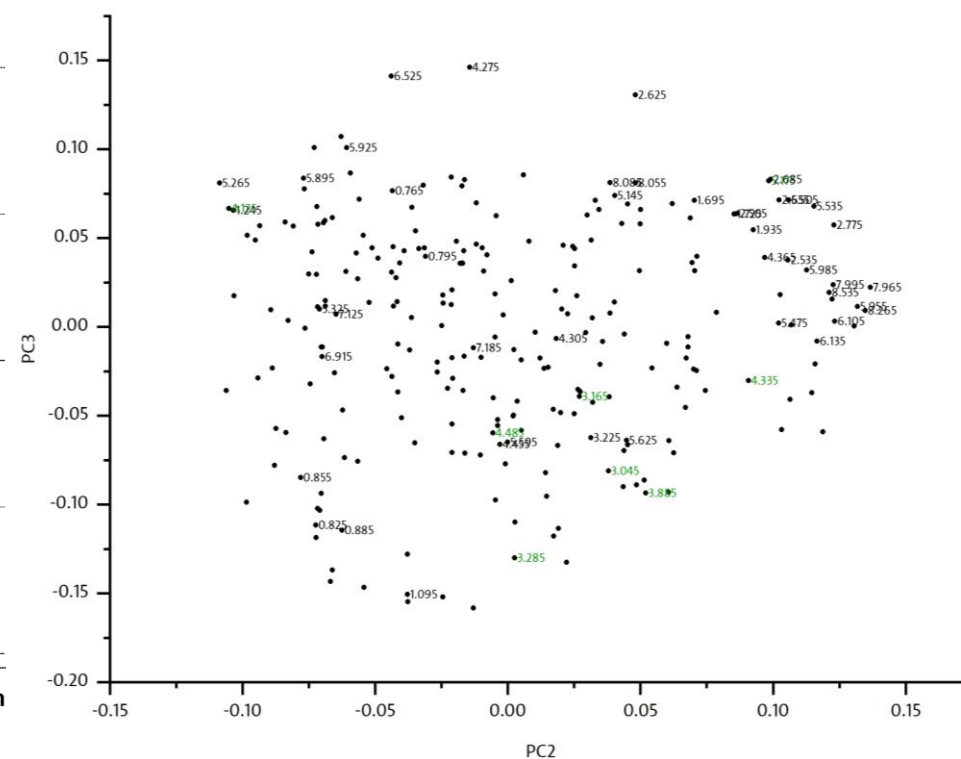
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# **NMR Metabolomics und die Analyse komplexer Gemische**

# Metabolomics-Studien: Artendifferenzierung Trüffel



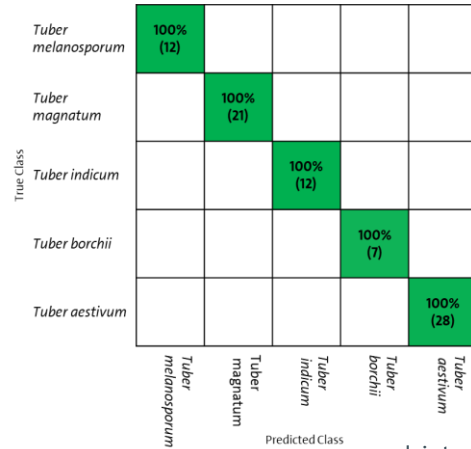
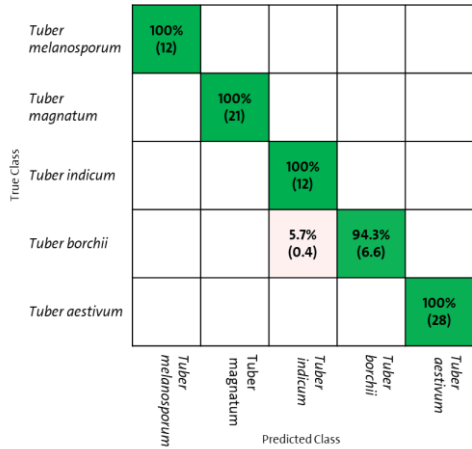
Hauptkomponentenanalyse (PCA): Loadings-Plot



# Klassifizierung: Support Vector Machine

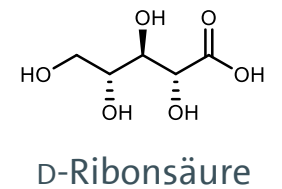
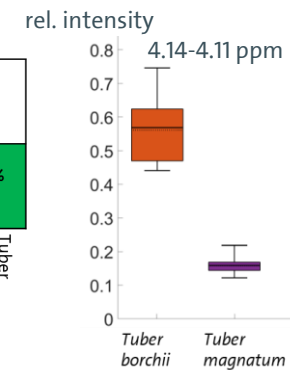
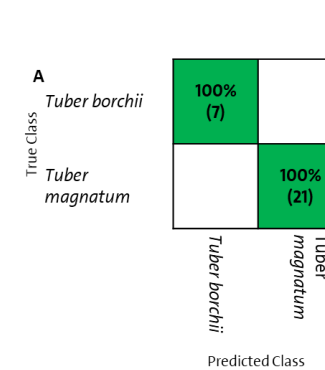
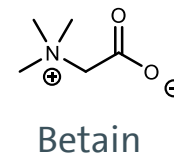
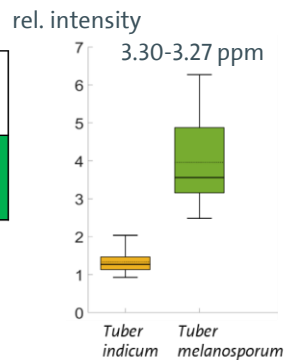
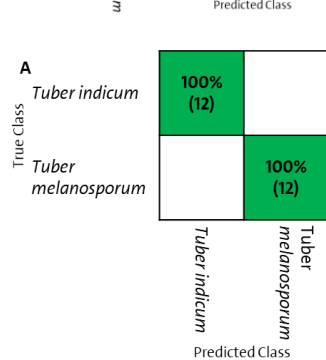
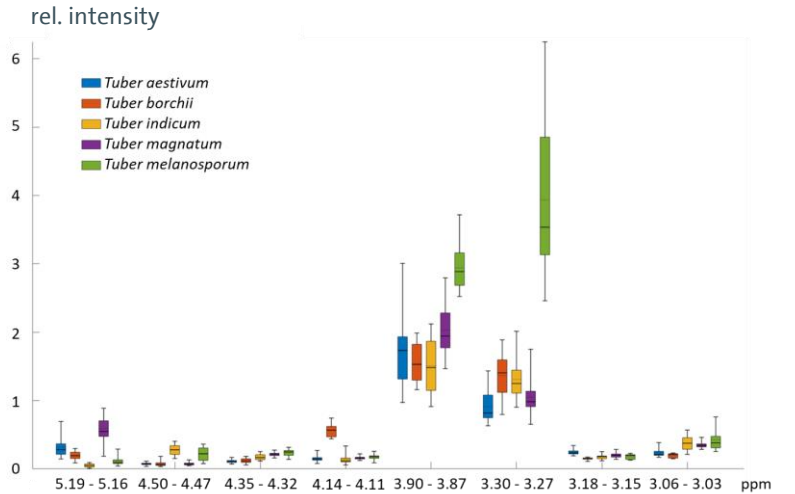
Konfusionsmatrix | 253 Features

Konfusionsmatrix | 8 Features



Boxplots | 8 Features nach Feature-Selektion

Alle 8 Features sind in einer one-way ANOVA signifikant (Bonferroni-Holm-Korrektur)  
5 Features sind in der Top-10



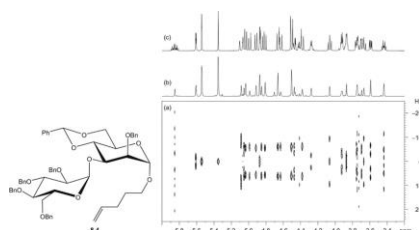
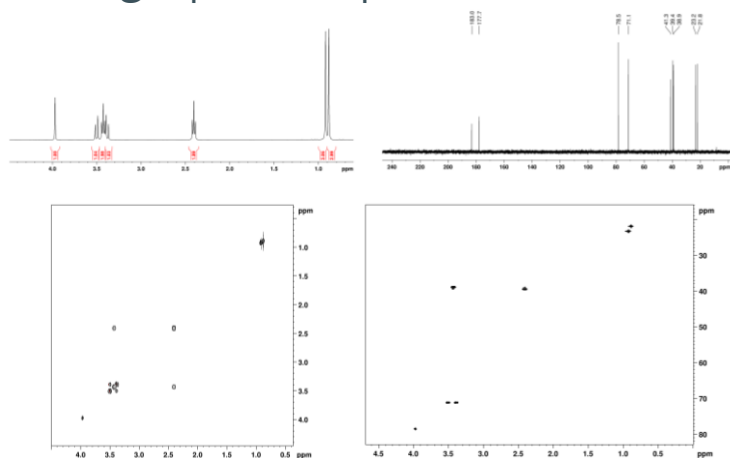
# Metabolite identifizieren | Gemische entschlüsseln

- Viele Tools und Datenbanken um eine Identifizierung zu unterstützen (Chenomx<sup>®</sup>, HMDB, BMRB, ...)

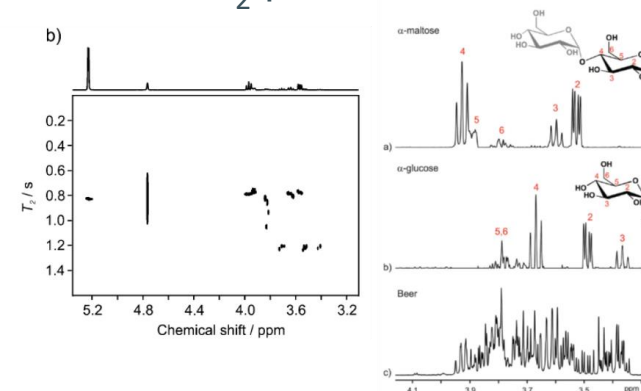
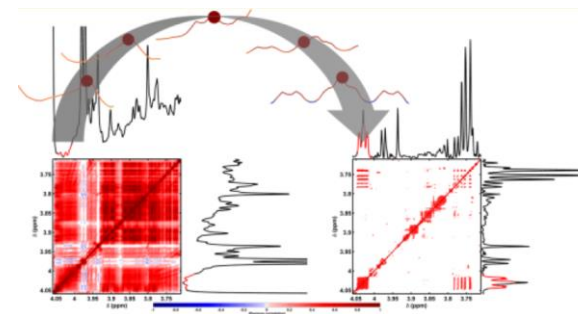
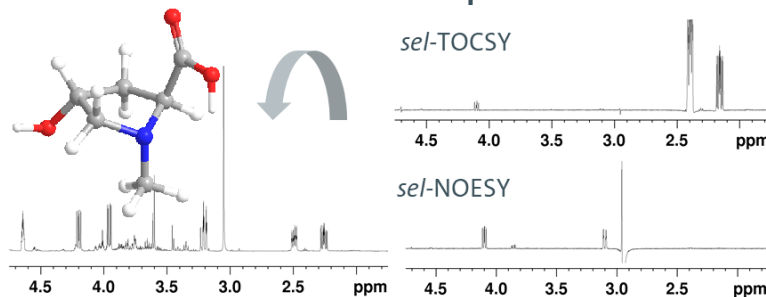
statistische Methoden  
 STOCSY | SHY | STORM

neue Experimente  
 z.B. REST<sub>2</sub> | SCALPEL

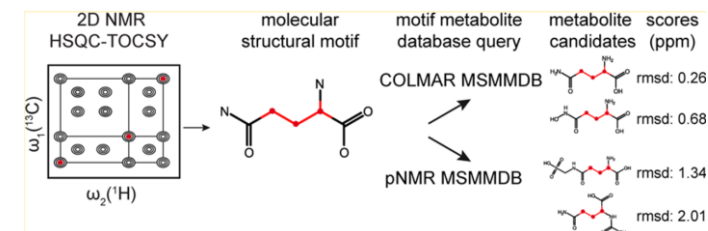
klassisch  
 big 5 | TOCSY | NOESY



J-res und selektive Experimente

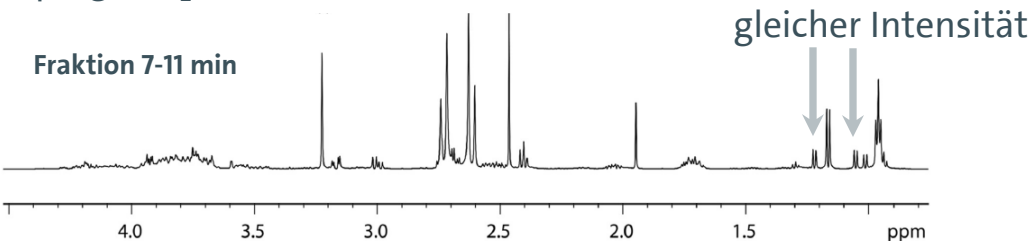


Strukturvorhersage | Neuronale Netze  
 SMART-NMR | COLMAR

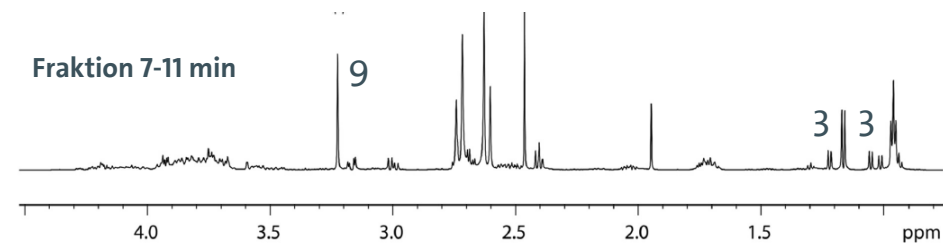
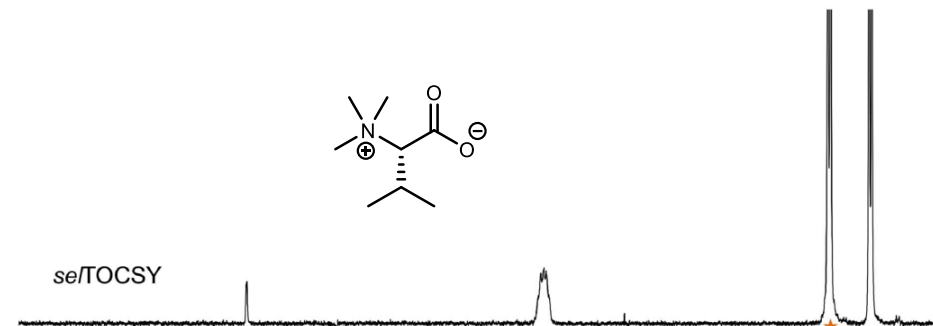
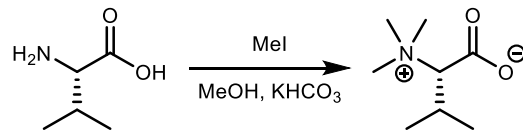
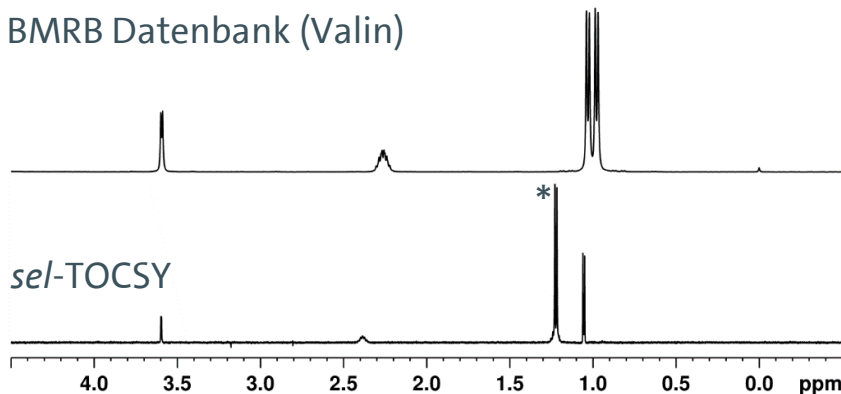


# Nur Datenbank?

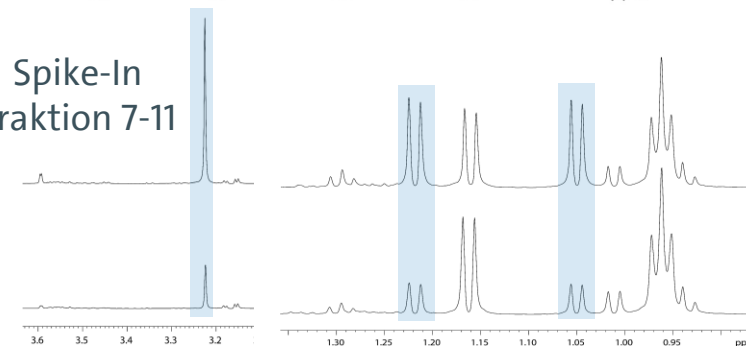
Spargel, H<sub>2</sub>O/MeOH Extract nach HPLC



BMRB Datenbank (Valin)

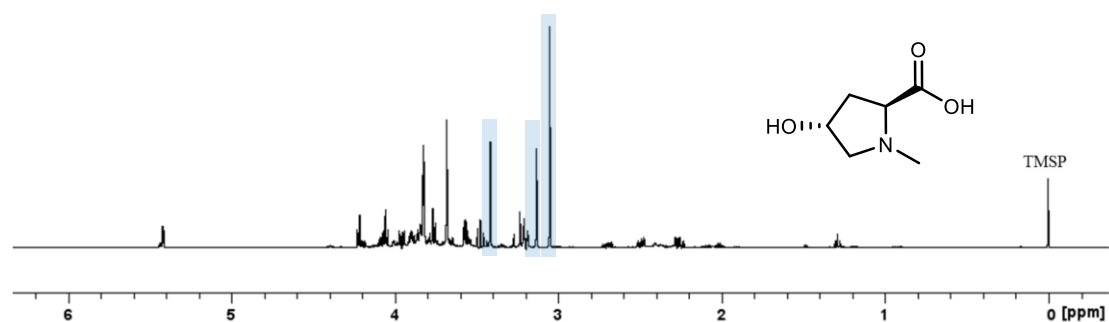


Spike-In  
Fraktion 7-11

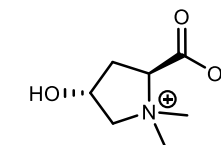
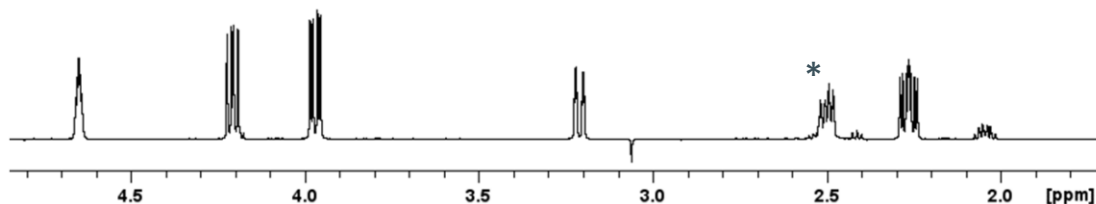


# NOTO und TONO

Cashew-Extrakt nach Säulenchromatographie  
3.3 mM *N*-Methyl-*trans*-4-hydroxy-L-prolin

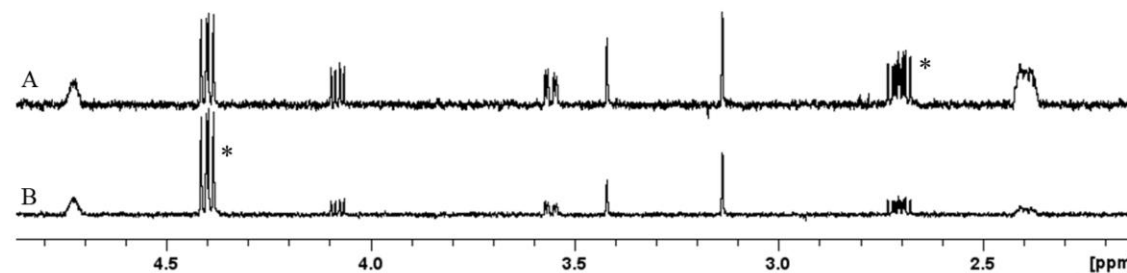


1D-*sel*-TOCSY-NOESY



*N,N*-Dimethyl-*trans*-4-hydroxy-L-prolin (Betonicin)

1D-*sel*-TOCSY-NOESY



S. Sciuto *et al.*, *Phytochemistry* **1983**, 22, 2311.

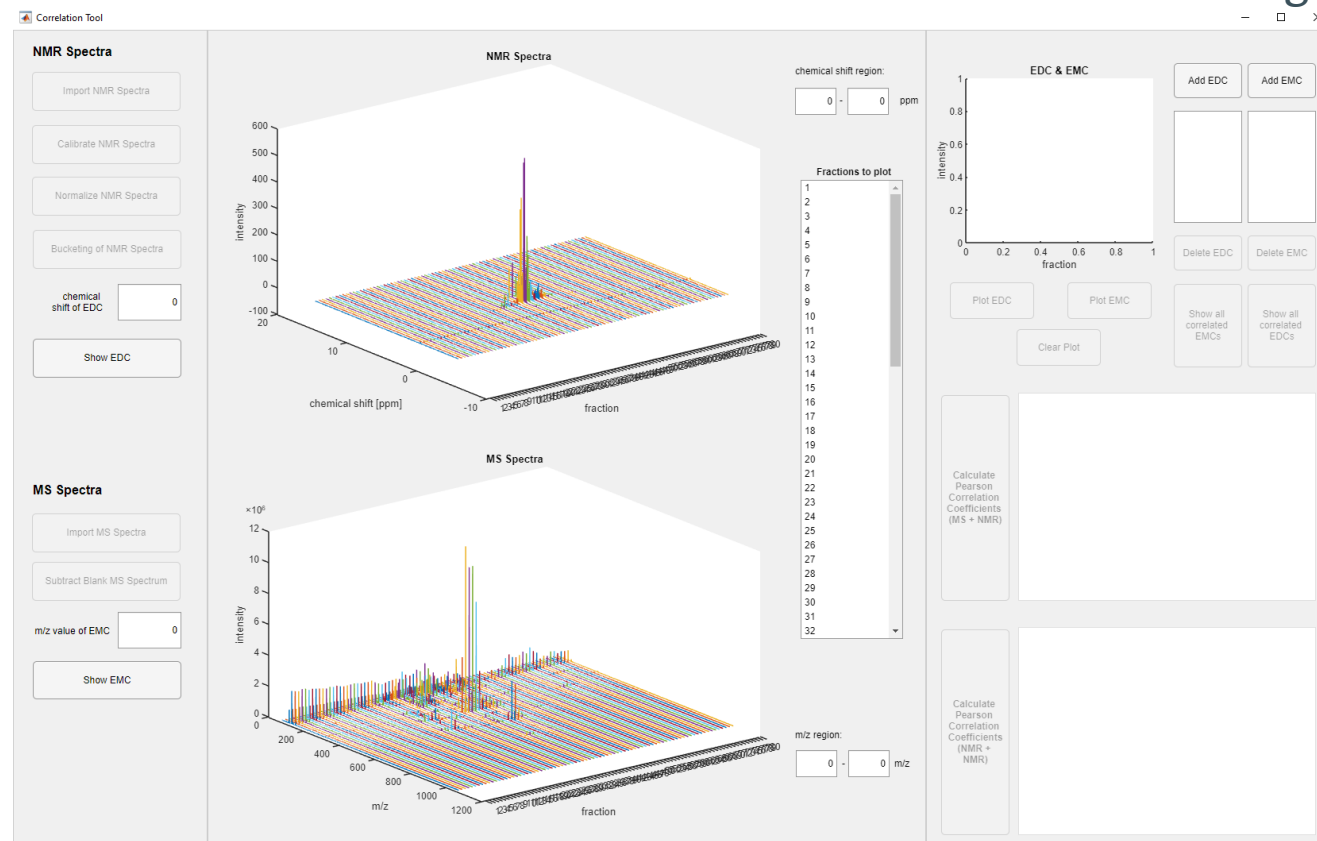
# SCORE-metabolite-ID

- Bereits angewendet
  - NMR/LC-MS *parallel dynamic spectroscopy* (NMR/LC-MS PDS) / Metaboliten
  - 3DCC (3-dimensional cross correlation) / Glycane
- MathLab®-App: Semiautomatische **C**orrelation analysis for **R**eliable **m**etabolite **I**dentification

Dai et al., *Anal. Chim. Acta* **2009**, 632, 221–228.

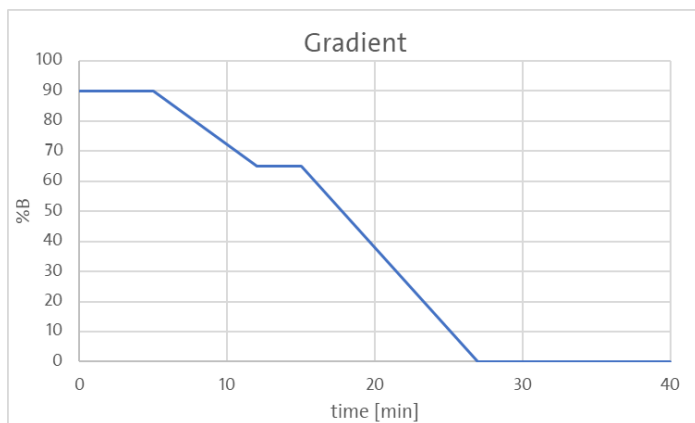
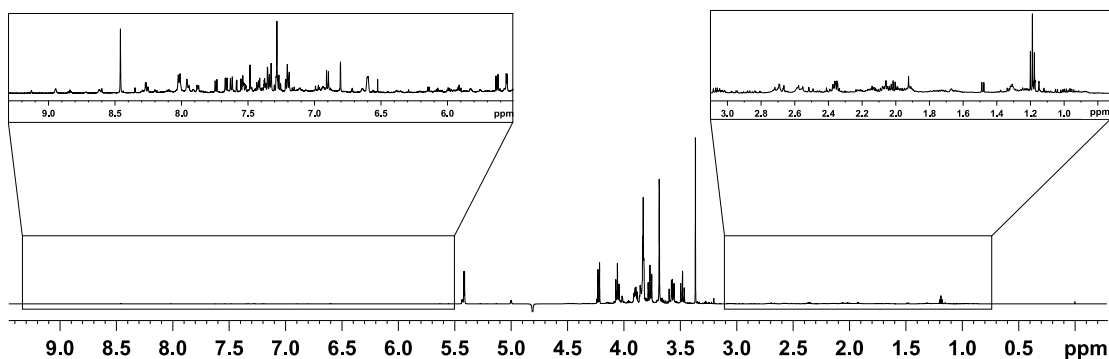
Behnken et al., *Anal. Bioanal. Chem.* **2012**, 404, 1427–1437.

EDC: Extracted Delta Chromatogram  
EMC: Extracted Mass Chromatogram

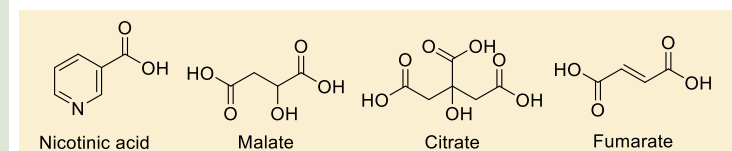
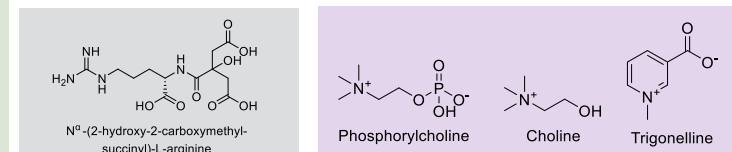
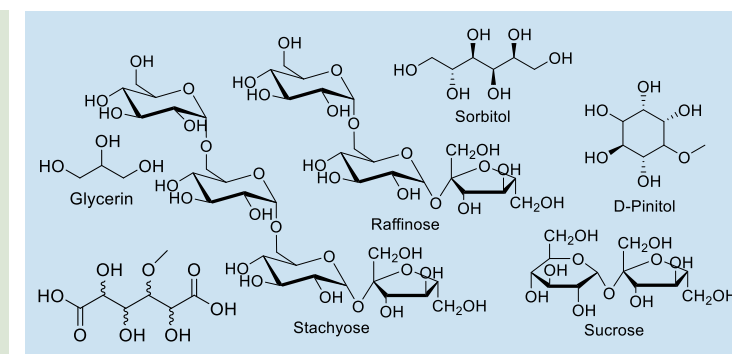
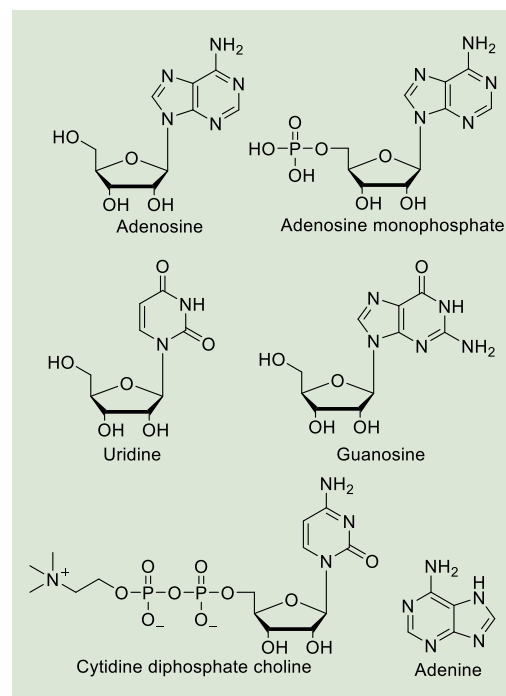
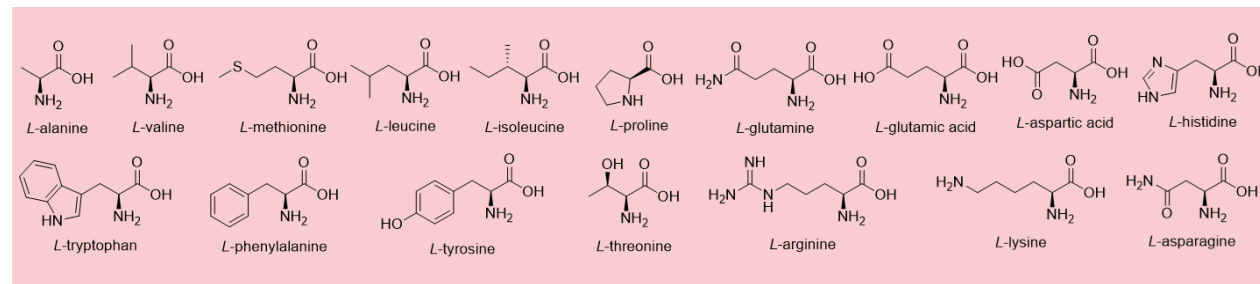




# Pinienkern-Extrakt



Fraktionierung unter  
 HILIC Bedingungen  
 polare Amino-säule  
 (A = H<sub>2</sub>O, B = ACN)  
 80 Fraktionen  
 NS 64 pro NMR-  
 Spektrum (27 h)





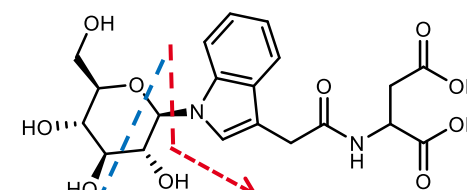
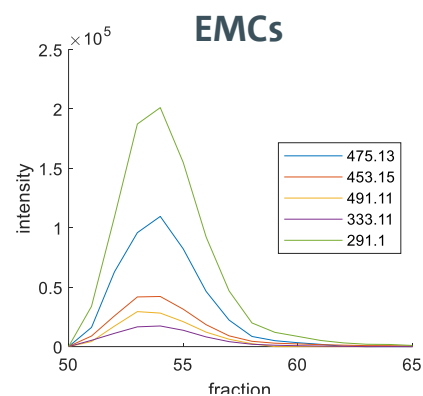
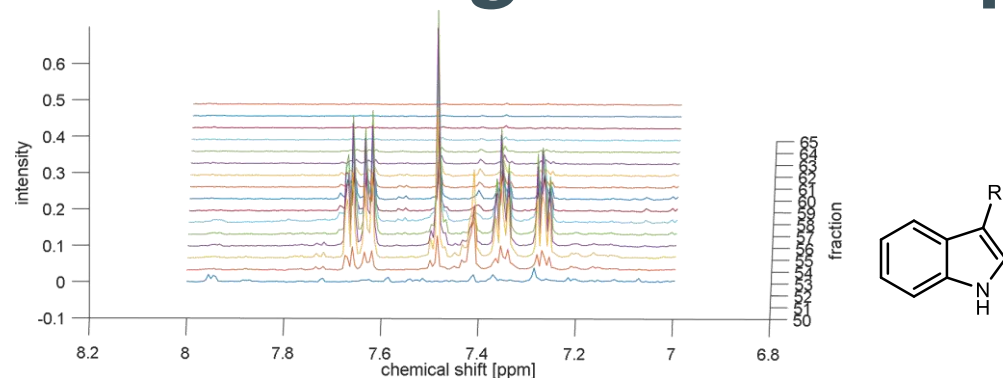
# Identifizierung von IAA-Asp-N-Glc

Mögliche Summenformeln:

$C_5H_{20}N_{14}O_{11}$ , DBE = 3,  $M_{ex} = 452.1436$

$C_{20}H_{24}N_2O_{10}$ , DBE = 10,  $M_{ex} = 452.1431$

$C_{18}H_{12}N_{16}$ , DBE = 21,  $M_{ex} = 452.1431$

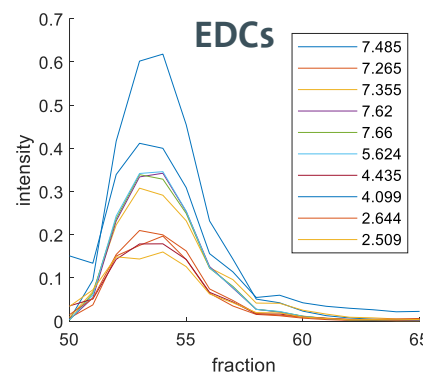


$m/z = 291.10$

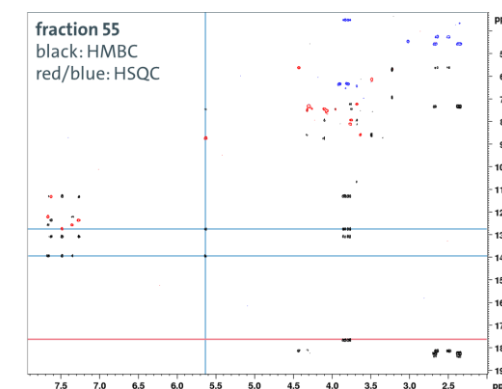
$m/z = 333.11$

korrelierende MS Signale zu NMR Signal bei 7.485 ppm

MS signal [m/z]	Pearson Correlation Coefficient	maximal intensity of MS signal	possible peak assignment
291.10	0.9930	201124	
475.13	0.9952	109562	[M+Na] <sup>+</sup>
453.15	0.9971	42182	[M+H] <sup>+</sup>
292.10	0.9955	41189	
178.99	0.9777	32023	
491.11	0.9962	29449	[M+K] <sup>+</sup>
476.14	0.9961	24595	[M+Na] <sup>+</sup> isotope
157.01	0.9875	21343	
333.11	0.9919	17367	
497.12	0.9886	14658	[M+2Na-H] <sup>+</sup>



Kenji Kai *et al.*, *Phytochemistry* **2007**, *68*, 2512–2522.



## Stephanie Watermann



Thorsten Mix

Ehemalige: Navid Shakiba, René Bachmann, Anke Bollen,  
Caroline Schmitt, Juliane Klare, Moritz Fölsing. M.Sc.: Till  
Steinmetz, Nathalie Holz

Collaborations (UHH):  
Markus Fischer, Marina Creydt, Henry Lösel  
Stephan Seifert, Soeren Wenck  
Christian Stark

Peter Schmieder

Studenten & Technische MA:  
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Julia Kröger, Julia Eichbaum  
Claudia Wontorra, Vera Priegnitz

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LA BILANCIA Trüffelhandels GmbH” und  
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