Ein Beitrag zur autarken Autoinjektor-Fähigkeit der Bundeswehr

AQbD-basierte HILIC-Methodenentwicklung zur Untersuchung obidoximhaltiger Antidot-Lösungen

Benedikt Sperber

Lt (SanOA)

09.10.2026

Inhalt

01

Kontext

02

Obidoxim

- Mögliches Verunreinigungsprofil
- Methodenentwicklung

03

Fazit

Nervenkampfstoffe im Kriegskontext: Syrien





Adopt a Revolution

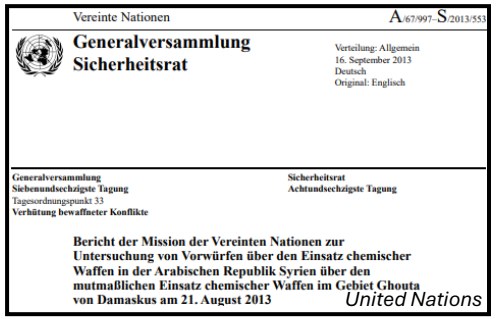
Kontext

Nervenkampfstoffe im Kriegskontext: Syrien





Adopt a Revolution


Vereinte Nationen A/67/997–S/2013/553

Generalversammlung
Sicherheitsrat

Verteilung: Allgemein
16. September 2013
Deutsch
Original: Englisch

Generalversammlung
Siebenundsechzigste Tagung
Tagesordnungspunkt 33
Verhütung bewaffneter Konflikte

Sicherheitsrat
Achtundsechzigste Tagung

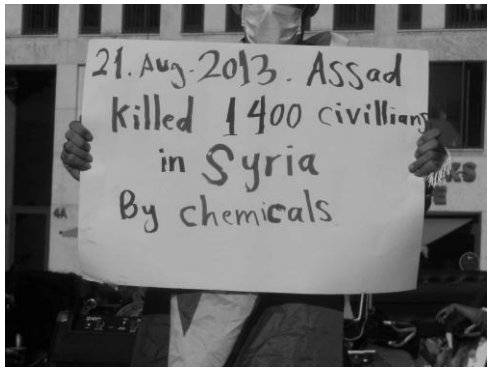
Bericht der Mission der Vereinten Nationen zur Untersuchung von Vorwürfen über den Einsatz chemischer Waffen in der Arabischen Republik Syrien über den mutmaßlichen Einsatz chemischer Waffen im Gebiet Ghouta von Damaskus am 21. August 2013

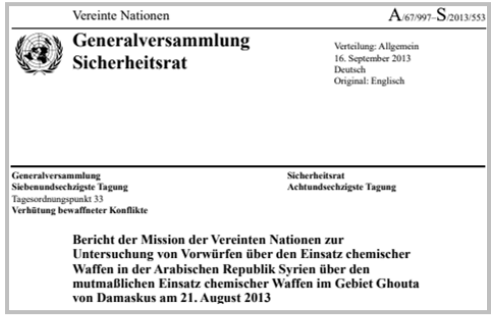
United Nations

Kontext

Nervenkampfstoffe im Kriegskontext: Syrien







Vereinte Nationen A/67/997-S/2013/553

Generalversammlung
Sicherheitsrat

Verteilung: Allgemein
16. September 2013
Deutsch
Original: Englisch

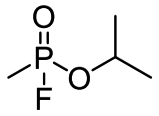
Generalversammlung
Siebenundsechzigste Tagung
Tagesordnungspunkt 33
Verhütung bewaffneter Konflikte

Sicherheitsrat
Achtundsechzigste Tagung

Bericht der Mission der Vereinten Nationen zur Untersuchung von Vorwürfen über den Einsatz chemischer Waffen in der Arabischen Republik Syrien über den mutmaßlichen Einsatz chemischer Waffen im Gebiet Ghouta von Damaskus am 21. August 2013

1) Angriff auf Ghouta 21.08.2013





Sarin



355 – 1709



3600 – 6000

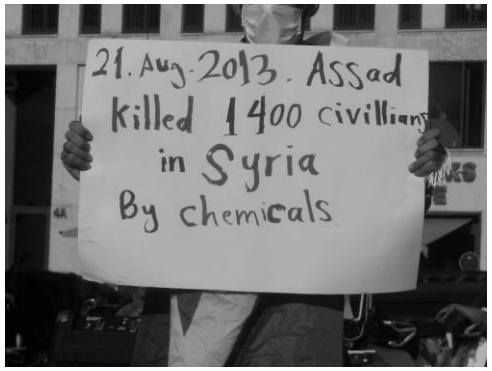


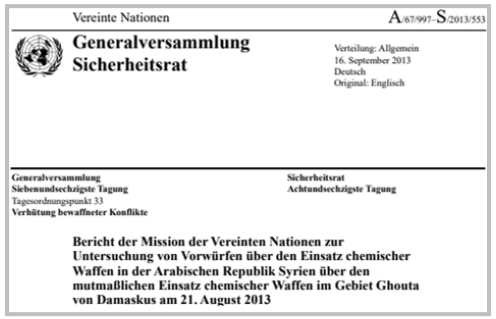
Bombadierung der Region Ghouta (Vororte von Damaskus) mit Boden-Boden Raketen durch die syrische Armee

Kontext

Nervenkampfstoffe im Kriegskontext: Syrien







Vereinte Nationen A/67/997–S/2013/553

Generalversammlung
Sicherheitsrat

Verteilung: Allgemein
16. September 2013
Deutsch
Original: Englisch

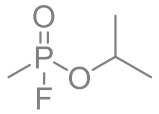
Generalversammlung
Siebenundsechzigste Tagung
Tagesordnungspunkt 33
Verhütung bewaffneter Konflikte

Sicherheitsrat
Achtundsechzigste Tagung

Bericht der Mission der Vereinten Nationen zur Untersuchung von Vorwürfen über den Einsatz chemischer Waffen in der Arabischen Republik Syrien über den mutmaßlichen Einsatz chemischer Waffen im Gebiet Ghouta von Damaskus am 21. August 2013

1) Angriff auf Ghouta 21.08.2013





Sarin



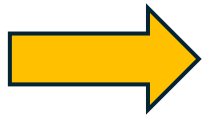
355 – 1709



3600 – 6000



Bombadierung der Region Ghouta (Vororte von Damaskus) mit Boden-Boden Raketen durch die syrische Armee

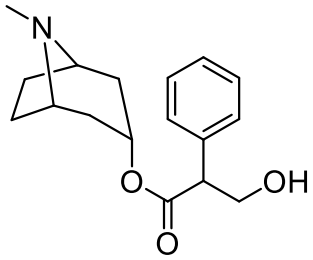


Beitritt Syriens zur Chemiewaffenkonvention (CWK, **14.09.2013**)

Kontext

Initiale Therapie bei Intoxikation: Autoinjektoren

1) **Atropin:** kompetitiver muskarinischer ACh-Rezeptorantagonist

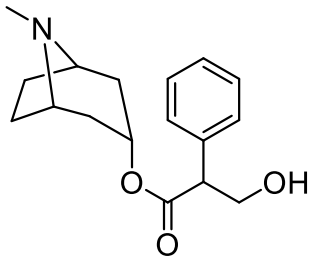


→**symptomatisches** Antidot

Kontext

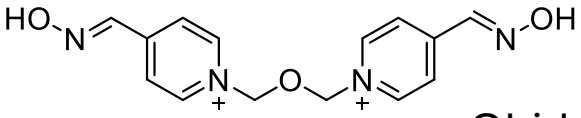
Initiale Therapie bei Intoxikation: Autoinjektoren

1) **Atropin:** kompetitiver muskarinischer ACh-Rezeptorantagonist

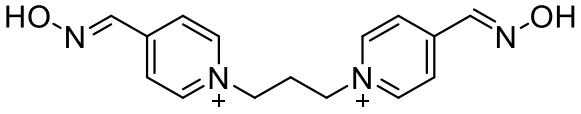


→**symptomatisches** Antidot

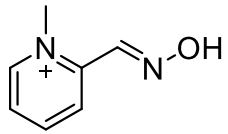
2) **Oxime:** Cholinesterase-Reaktivatoren



Obidoxim



Trimedoxim



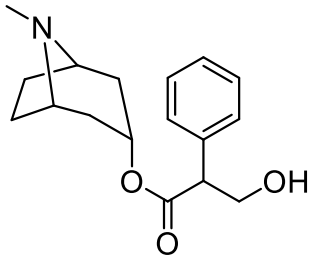
→**kausales** Antidot

Pralidoxim

Kontext

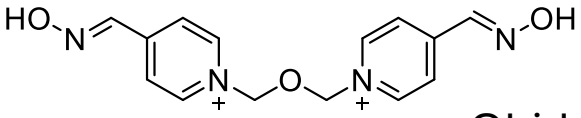
Initiale Therapie bei Intoxikation: Autoinjektoren

1) **Atropin:** kompetitiver muskarinischer ACh-Rezeptorantagonist

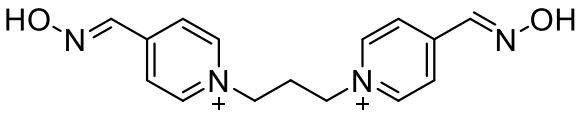


→**symptomatisches** Antidot

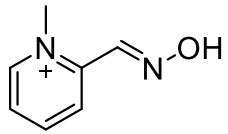
2) **Oxime:** Cholinesterase-Reaktivatoren



Obidoxim



Trimedoxim



→**kausales** Antidot

Pralidoxim

3) Autoinjektoren der BW



AtroPen®
Atropin

ATOX II
ComboPen®
Atropin
Obidoxim

Meridian
MEDICAL TECHNOLOGIES
Manufacturing More Tomorrows™

Kontext

Versorgungslücke Autoinjektor



ATOX II ComboPen®

- Keine Zulassung in DE
- Keine Herstellung mehr

Kontext

Versorgungslücke Autoinjektor





Hersteller?

ATOX II ComboPen®

- Keine Zulassung in DE
- Keine Herstellung mehr

→Beschaffung von Alternativen?

ATNAA

Atropin

Pralidoxim

Kontext

Versorgungslücke Autoinjektor



ATOX II ComboPen®

- Keine Zulassung in DE
- Keine Herstellung mehr
  →Beschaffung von Alternativen?



ATNAA
Atropin
Pralidoxim

Hersteller? → Meridian MEDICAL TECHNOLOGIES
Manufacturing More Tomorrows™

Kontext

Versorgungslücke Autoinjektor



ATOX II ComboPen®



ATNAA
Atropin
Pralidoxim

Hersteller?

Meridian
MEDICAL TECHNOLOGIES
Manufacturing More Tomorrows™

U.S. FOOD & DRUG ADMINISTRATION

Kansas City District Office
8050 Marshall Drive - Suite 205
Lenexa, Kansas 66214-1524
913-495-5100

September 5, 2017

WARNING LETTER

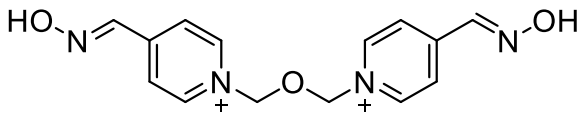
Ref: CMS Case: 525881
DELIVERY VIA UPS

Mr. Thomas E. Handel
President and General Manager
Meridian Medical Technologies, Inc., a Pfizer Company
6350 Stevens Forest Road, Suite 301
Columbia, MD 21046

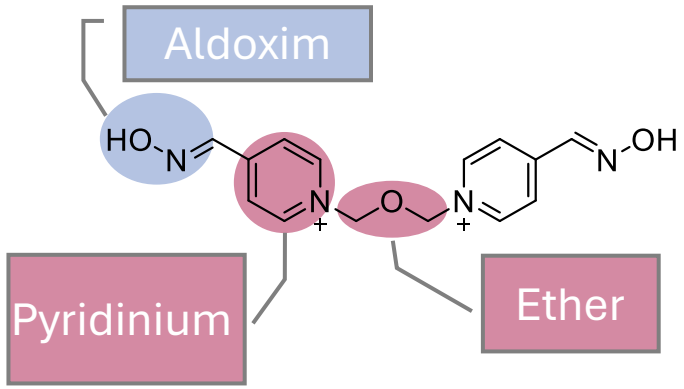
- Keine Zulassung in DE
- Keine Herstellung mehr
  →Beschaffung von Alternativen?

Kontext

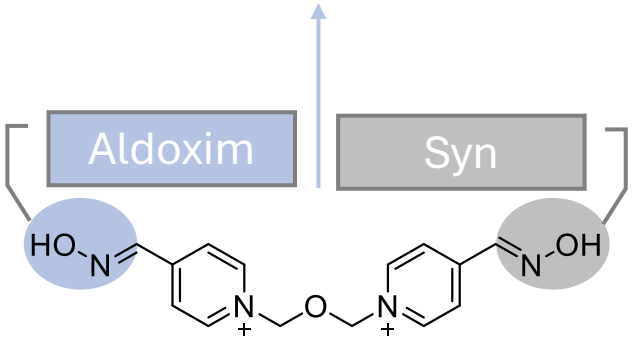
Obidoxim: Der Analyt und mögliche Abbauprodukte



Obidoxim: Der Analyt und mögliche Abbauprodukte

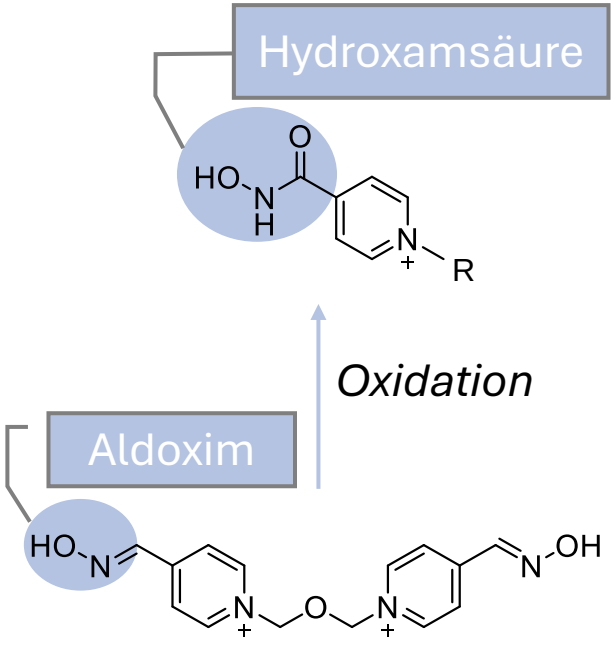


Obidoxim: Der Analyt und mögliche Abbauprodukte



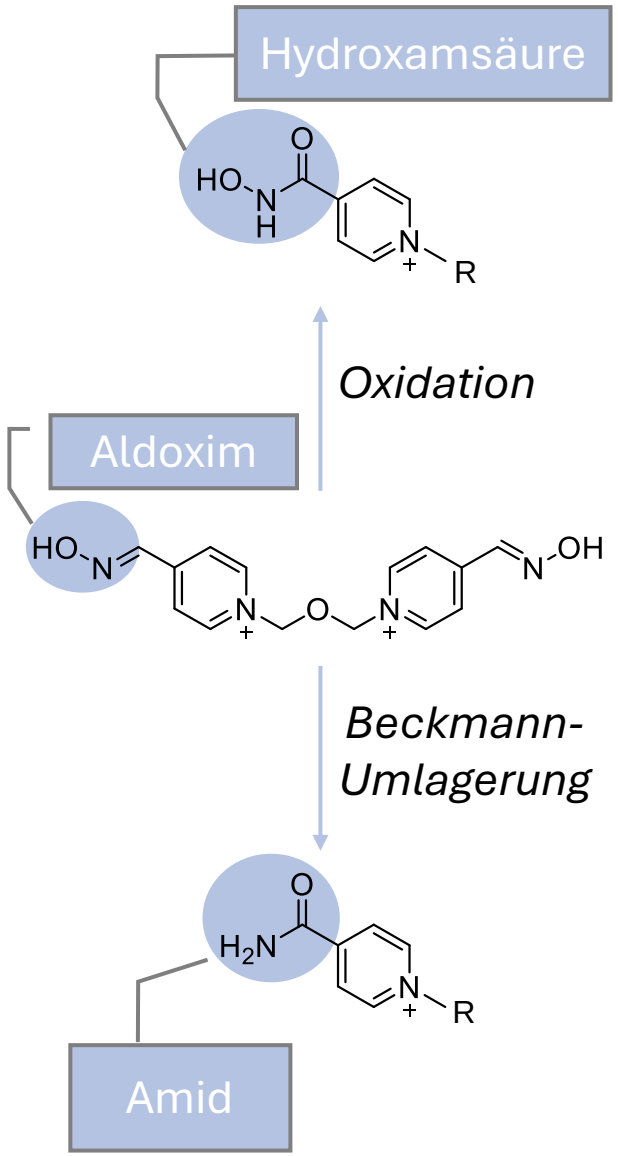
(1) S. Rubnov, I. Shats, D. Levy, S. Amisar, H. Schneider, Autocatalytic degradation and stability of obidoxime, Journal of pharmacy and pharmacology, 51 (1999) 9-14. (2) I. Christenson, Hydrolysis of bis (4-hydroxyiminomethyl-1-pyridiniomethyl) ether dichloride (Toxogonin). II. Kinetics and equilibrium in acidic solution, Acta pharmaceutica Suecica, 5 (1968) 249-262.
(3) P. Eyer, W. Hell, A. Kawan, H. Klehr, Studies on the decomposition of the oxime HI 6 in aqueous solution, Arch Toxicol, 59 (1986) 266–271.

Obidoxim: Der Analyt und mögliche Abbauprodukte



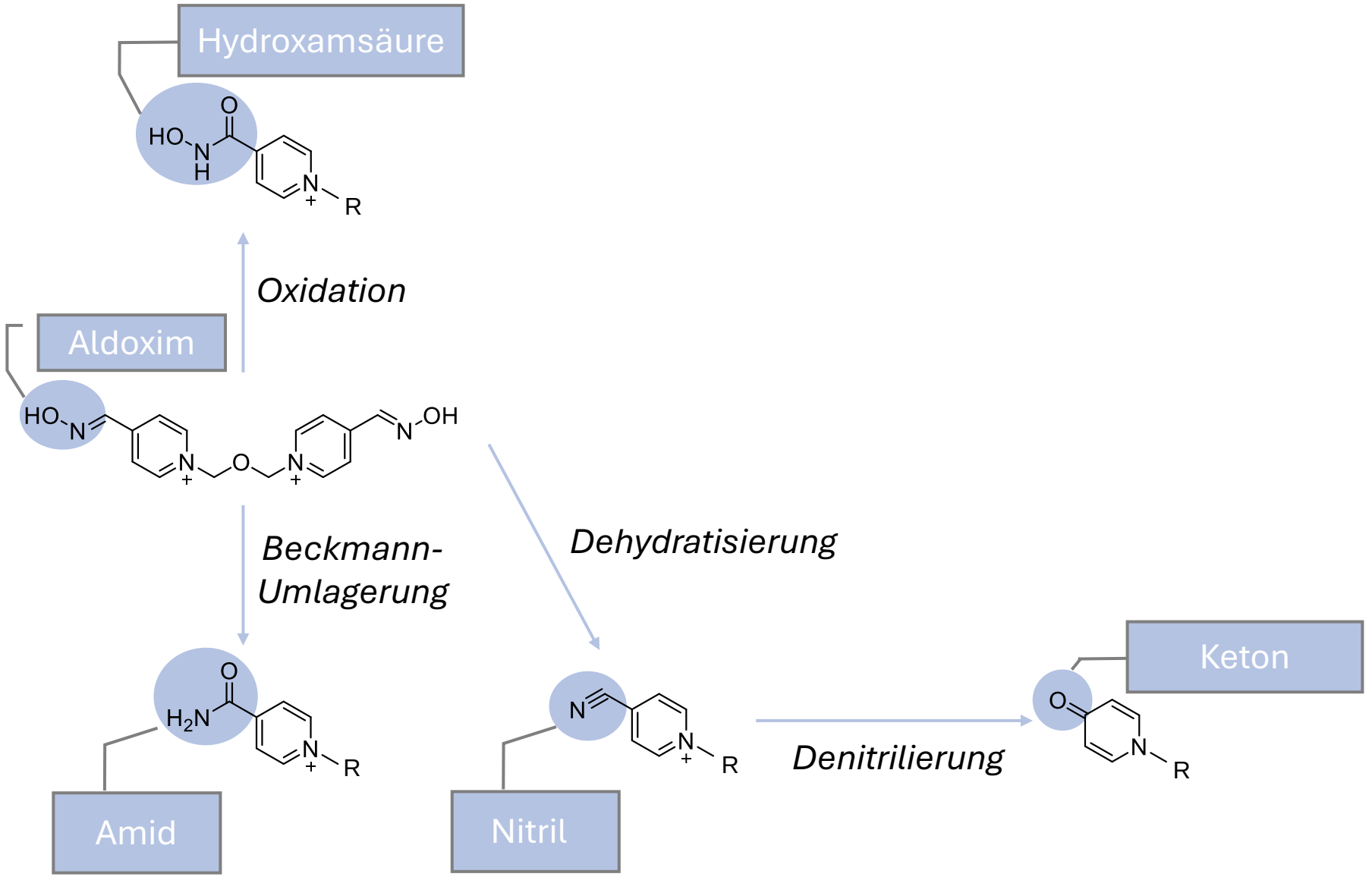
(1) S. Rubnov, I. Shats, D. Levy, S. Amisar, H. Schneider, Autocatalytic degradation and stability of obidoxime, Journal of pharmacy and pharmacology, 51 (1999) 9-14. (2) I. Christenson, Hydrolysis of bis (4-hydroxyiminomethyl-1-pyridiniomethyl) ether dichloride (Toxogonin). II. Kinetics and equilibrium in acidic solution, Acta pharmaceutica Suecica, 5 (1968) 249-262.
(3) P. Eyer, W. Hell, A. Kawan, H. Klehr, Studies on the decomposition of the oxime HI 6 in aqueous solution, Arch Toxicol, 59 (1986) 266–271.

Obidoxim: Der Analyt und mögliche Abbauprodukte



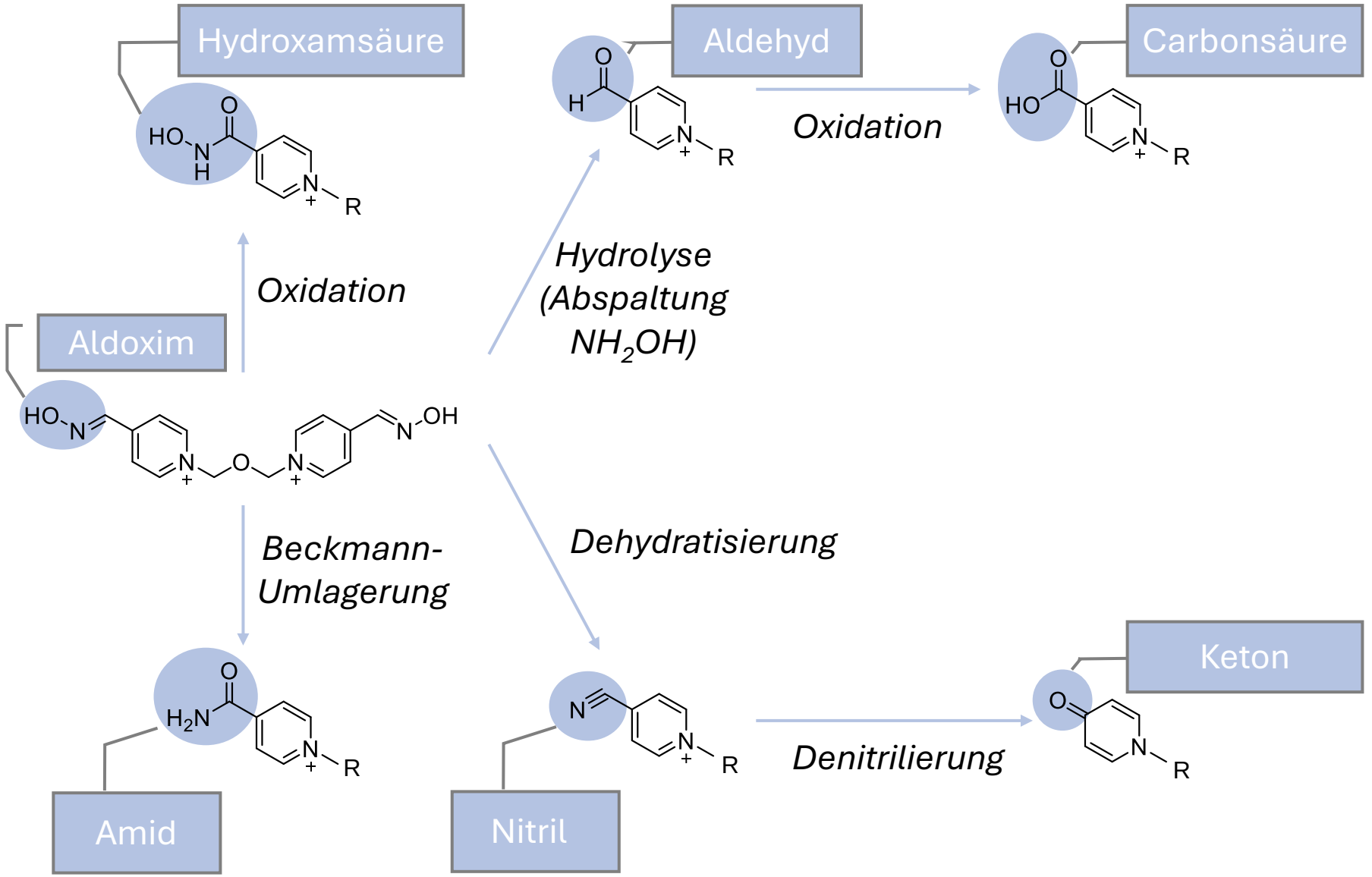
(1) S. Rubnov, I. Shats, D. Levy, S. Amisar, H. Schneider, Autocatalytic degradation and stability of obidoxime, Journal of pharmacy and pharmacology, 51 (1999) 9-14. (2) I. Christenson, Hydrolysis of bis (4-hydroxyiminomethyl-1-pyridiniomethyl) ether dichloride (Toxogonin). II. Kinetics and equilibrium in acidic solution, Acta pharmaceutica Suecica, 5 (1968) 249-262.
(3) P. Eyer, W. Hell, A. Kawan, H. Klehr, Studies on the decomposition of the oxime HI 6 in aqueous solution, Arch Toxicol, 59 (1986) 266–271.

Obidoxim: Der Analyt und mögliche Abbauprodukte



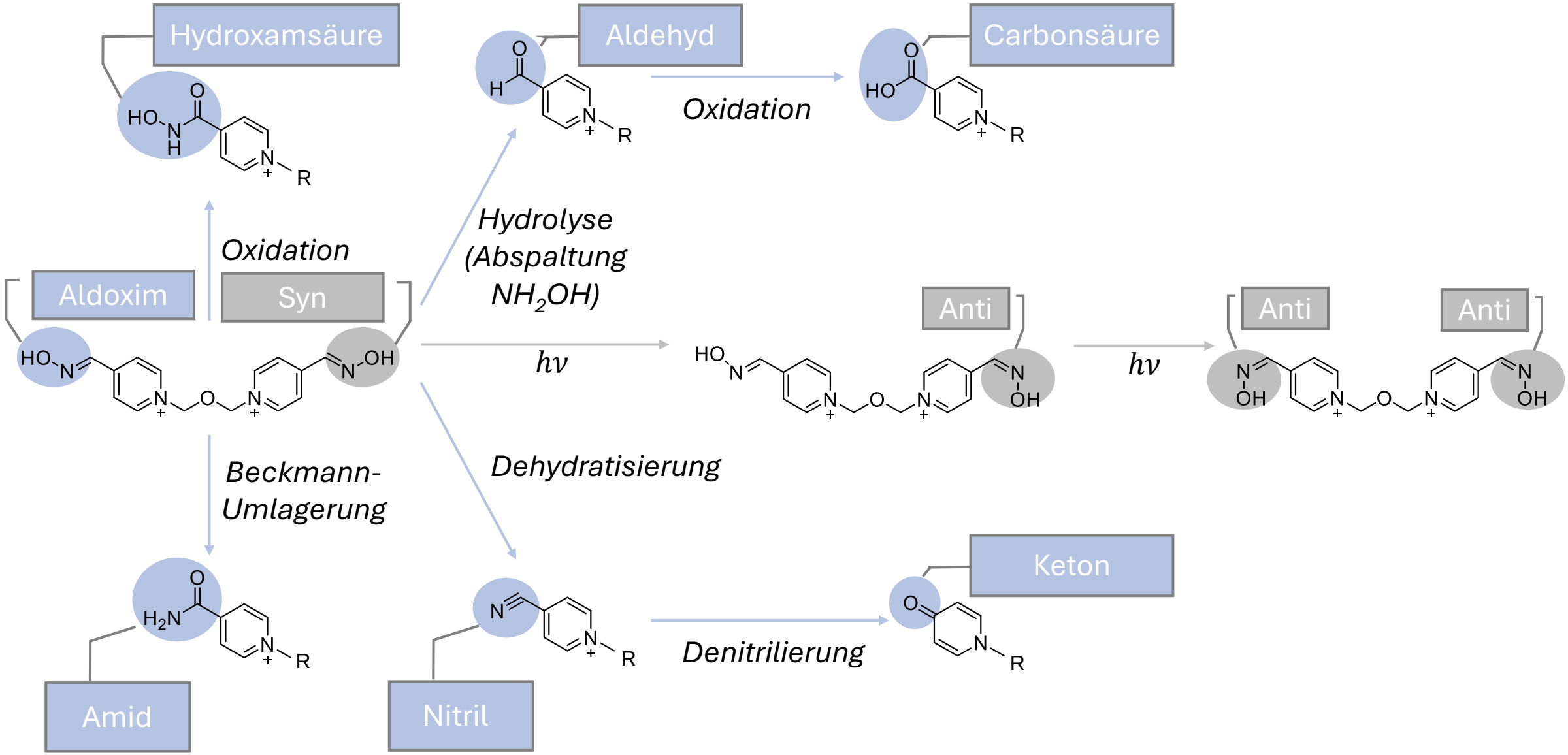
(1) S. Rubnov, I. Shats, D. Levy, S. Amisar, H. Schneider, Autocatalytic degradation and stability of obidoxime, Journal of pharmacy and pharmacology, 51 (1999) 9-14. (2) I. Christenson, Hydrolysis of bis (4-hydroxyiminomethyl-1-pyridiniomethyl) ether dichloride (Toxogonin). II. Kinetics and equilibrium in acidic solution, Acta pharmaceutica Suecica, 5 (1968) 249-262.
(3) P. Eyer, W. Hell, A. Kawan, H. Klehr, Studies on the decomposition of the oxime HI 6 in aqueous solution, Arch Toxicol, 59 (1986) 266–271.

Obidoxim: Der Analyt und mögliche Abbauprodukte



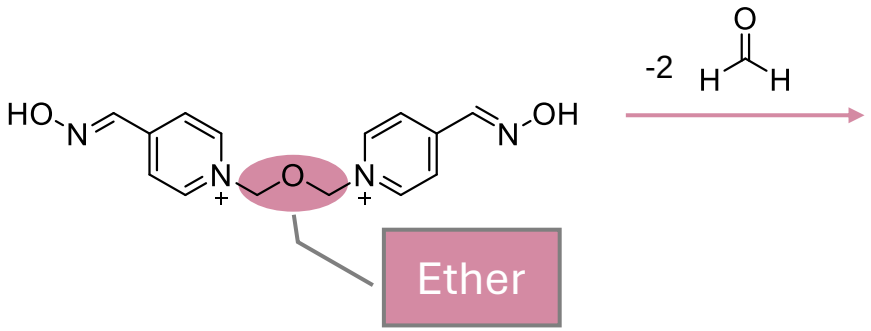
(1) S. Rubnov, I. Shats, D. Levy, S. Amisar, H. Schneider, Autocatalytic degradation and stability of obidoxime, Journal of pharmacy and pharmacology, 51 (1999) 9-14. (2) I. Christenson, Hydrolysis of bis (4-hydroxyiminomethyl-1-pyridiniomethyl) ether dichloride (Toxogonin). II. Kinetics and equilibrium in acidic solution, Acta pharmaceutica Suecica, 5 (1968) 249-262.
(3) P. Eyer, W. Hell, A. Kawan, H. Klehr, Studies on the decomposition of the oxime HI 6 in aqueous solution, Arch Toxicol, 59 (1986) 266–271.

Obidoxim: Der Analyt und mögliche Abbauprodukte

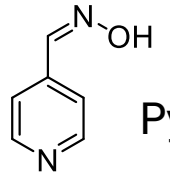


(1) S. Rubnov, I. Shats, D. Levy, S. Amisar, H. Schneider, Autocatalytic degradation and stability of obidoxime, Journal of pharmacy and pharmacology, 51 (1999) 9-14. (2) I. Christenson, Hydrolysis of bis (4-hydroxyiminomethyl-1-pyridiniomethyl) ether dichloride (Toxogonin). II. Kinetics and equilibrium in acidic solution, Acta pharmaceutica Suecica, 5 (1968) 249-262.
(3) P. Eyer, W. Hell, A. Kawan, H. Klehr, Studies on the decomposition of the oxime HI 6 in aqueous solution, Arch Toxicol, 59 (1986) 266–271. (4) R. Bogan, F. Worek, M. Koller, B. Klaubert, Photostability of antidotal oxime HI-6, impact on drug development, Drug Testing and Analysis, 4 (2012) 208–214.

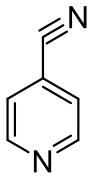
Obidoxim: Der Analyt und mögliche Abbauprodukte



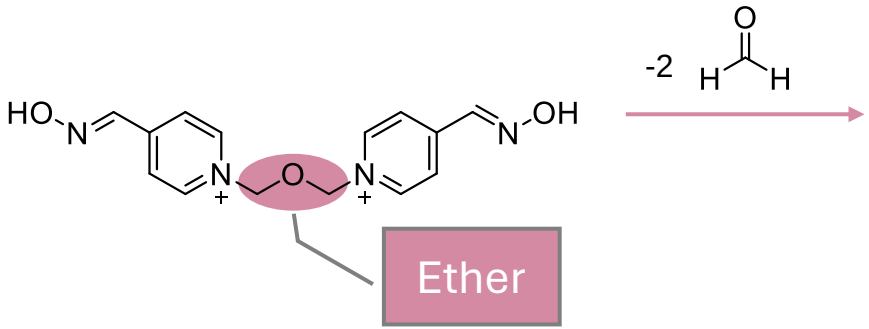
Obidoxim: Der Analyt und mögliche Abbauprodukte



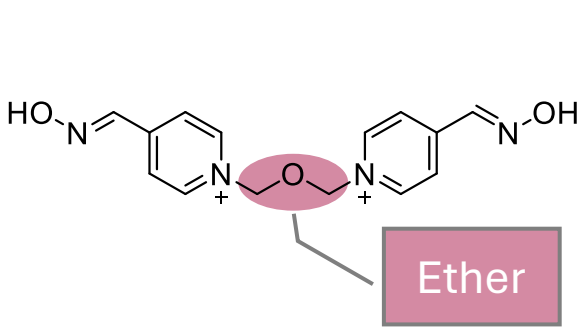
Pyridin-4-Aldoxim (Edukt)

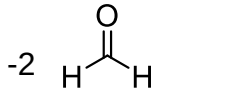


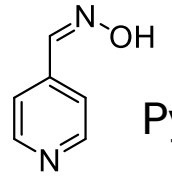
Pyridin-4-carbonitril



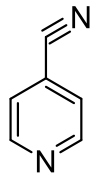
Obidoxim: Der Analyt und mögliche Abbauprodukte



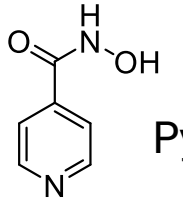




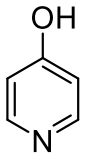
Pyridin-4-Aldoxim (Edukt)



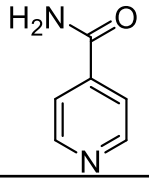
Pyridin-4-carbonitril



Pyridin-4-hydroxamsäure

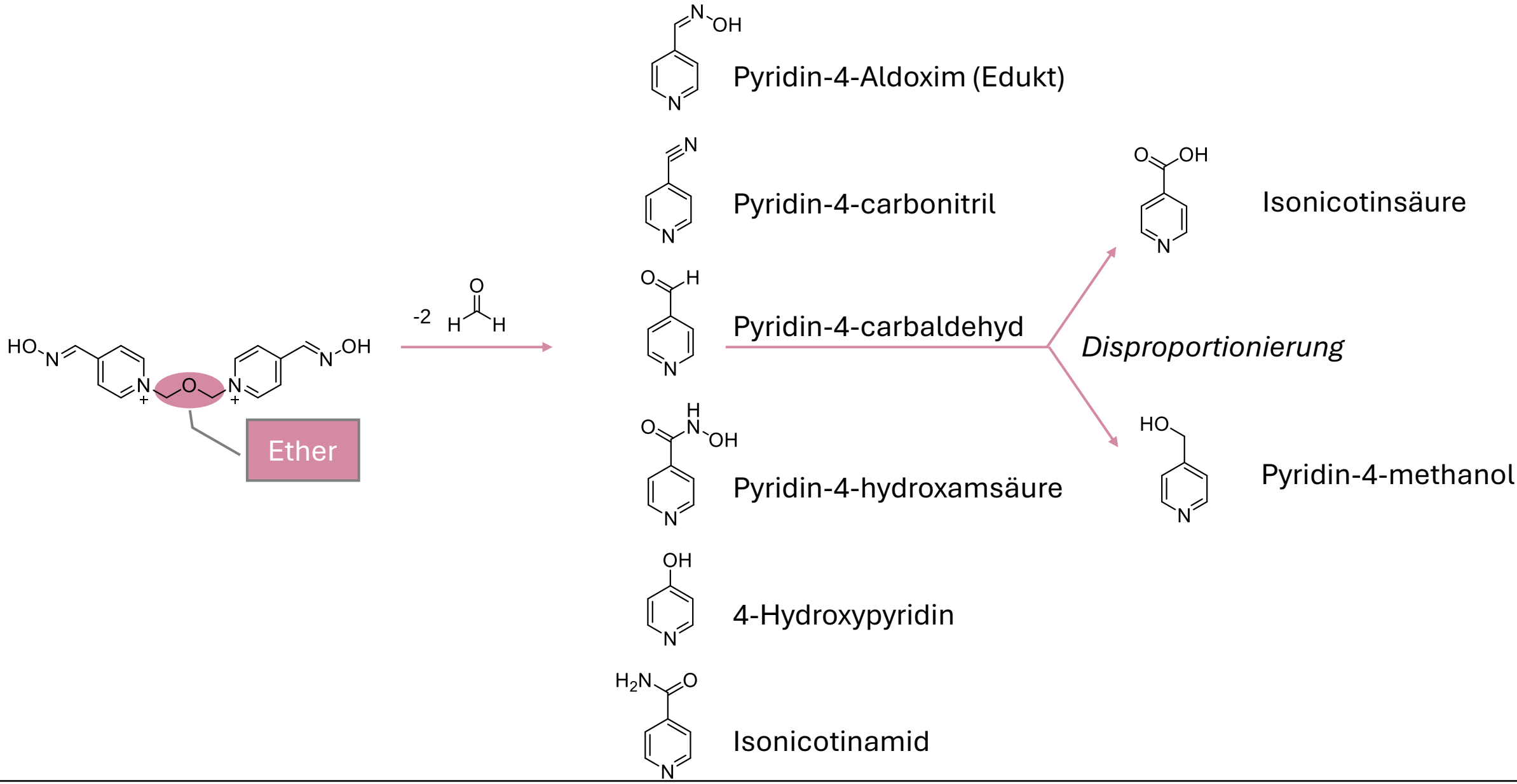


4-Hydroxypyridin



Isonicotinamid

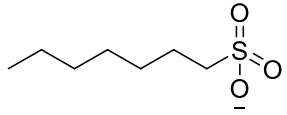
Obidoxim: Der Analyt und mögliche Abbauprodukte

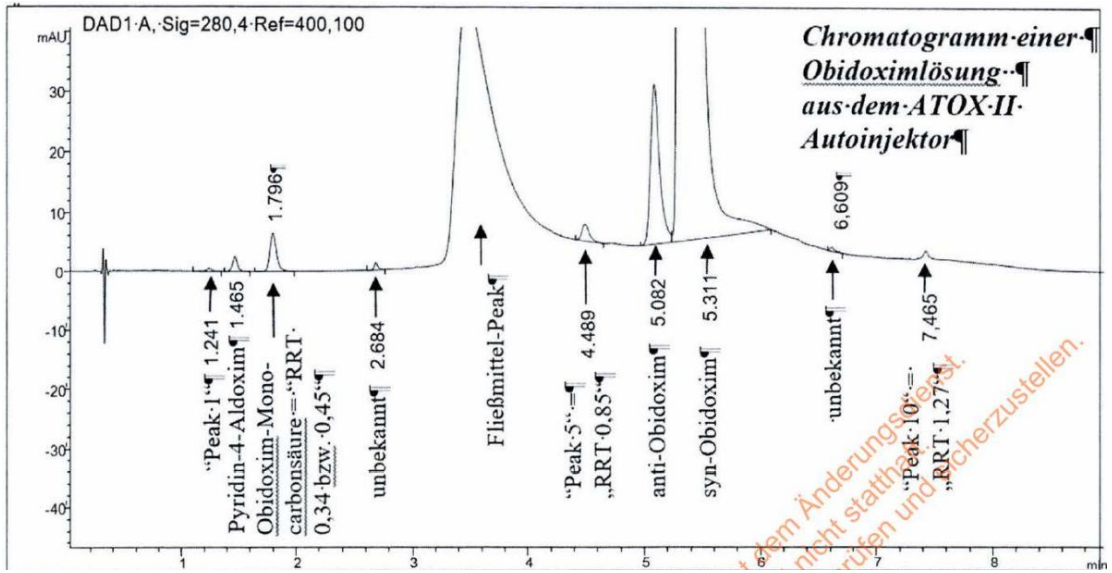


Obidoxim: Analytische Ansätze

1)Ionenpaarchromatographie auf C8/C18-Phasen

Ionenpaarreagenz



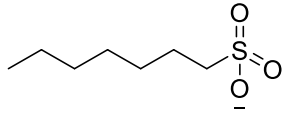


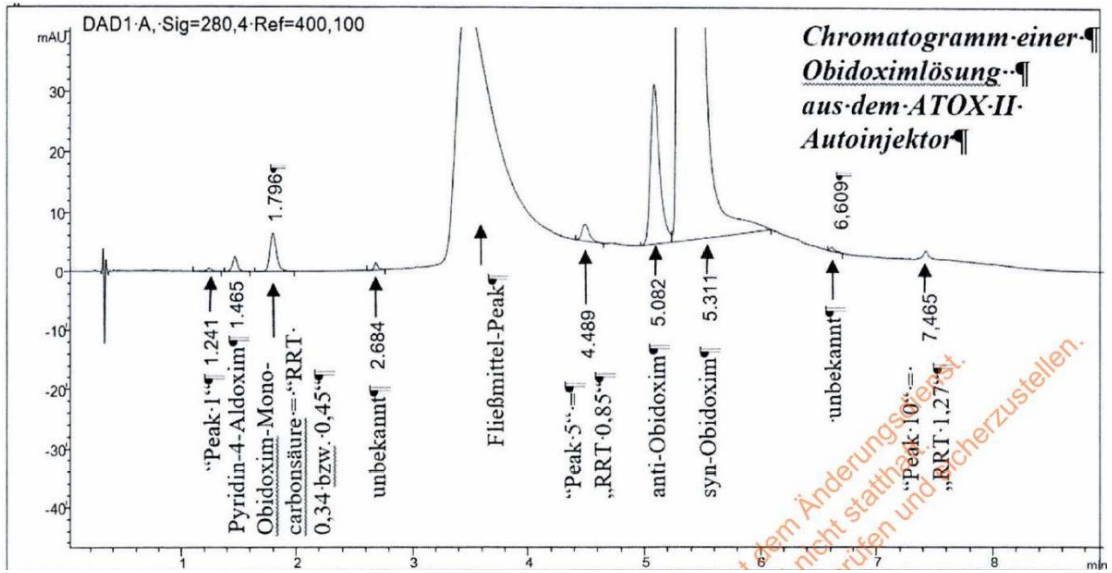
→nicht LC-MS tauglich

Obidoxim: Analytische Ansätze

1)Ionenpaarchromatographie auf C8/C18-Phasen

Ionenpaarreagenz

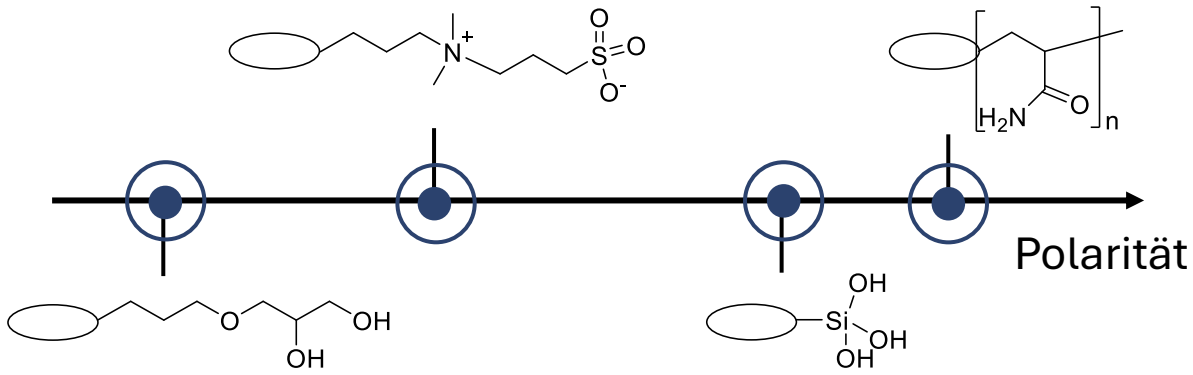




→nicht LC-MS tauglich

2) HILIC

- Retention für stark polare Substanzen

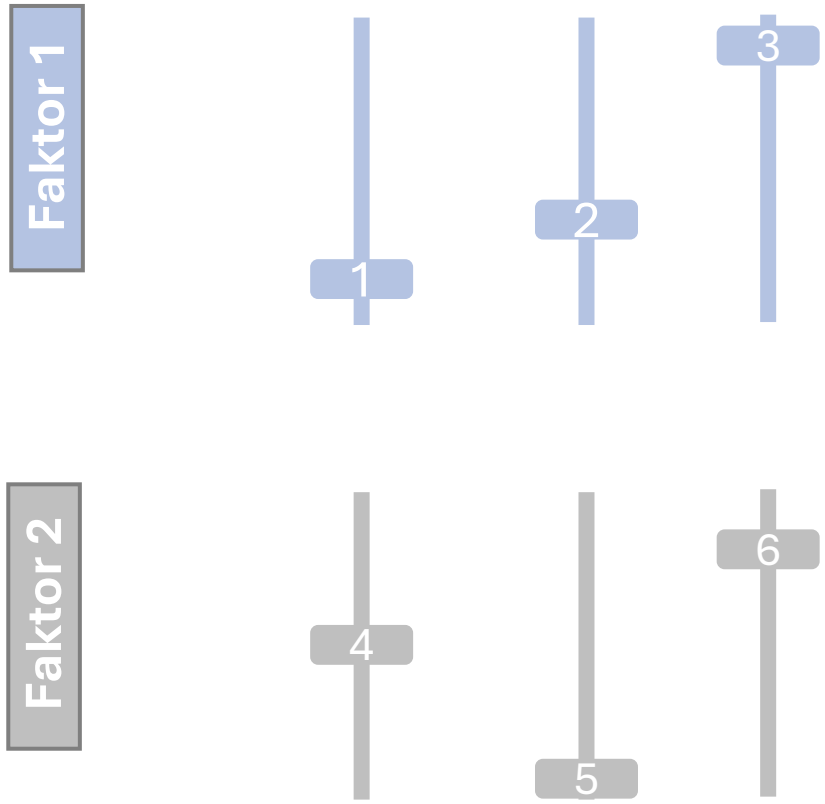


→LC-MS tauglich

(1) McCalley, D. V. Understanding and manipulating the separation in hydrophilic interaction liquid chromatography. J Chromatogr A 2017, 1523, 49-71. DOI: 10.1016/j.chroma.2017.06.026 From NLM Medline. (2) ThermoFisherScientific HILIC Separations: Pracitcal guide to HILIC mechanisms, method development and troubleshooting.

One factor at a time oder Analytical Quality by design?

One Factor at a time

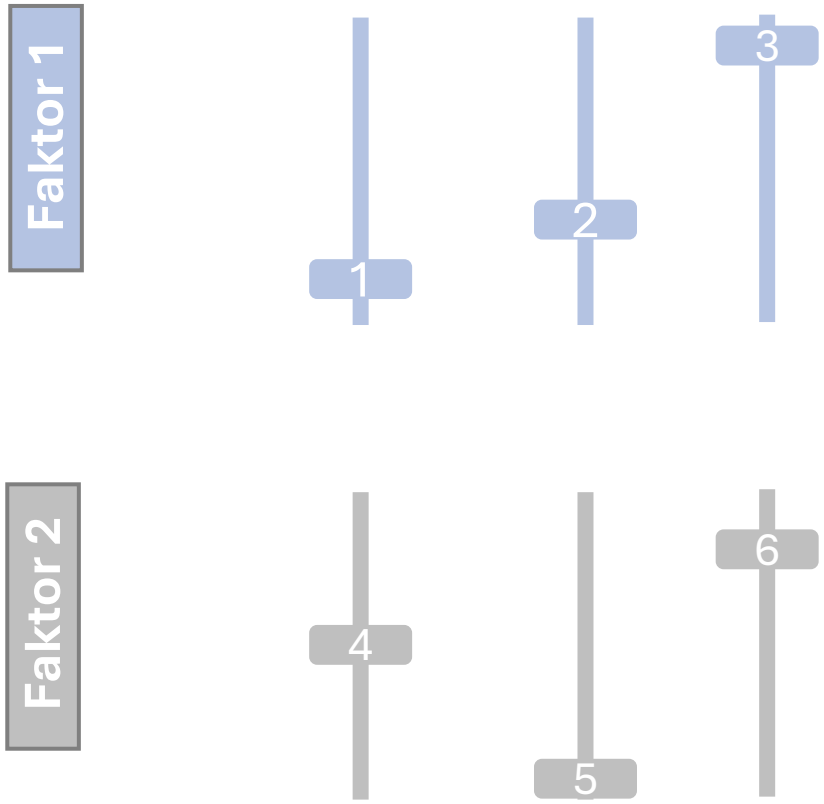


→fehlendes Methodenverständnis / Robustheit

(1) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q14: Analytical Procedure Development, (2023). (2) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q9: Quality risk management, (2025).
(3) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised guideline Q8(R2): Pharmaceutical development, (2009). (4) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q10: Pharmaceutical quality system, (2008).

One factor at a time oder Analytical Quality by design?

One Factor at a time

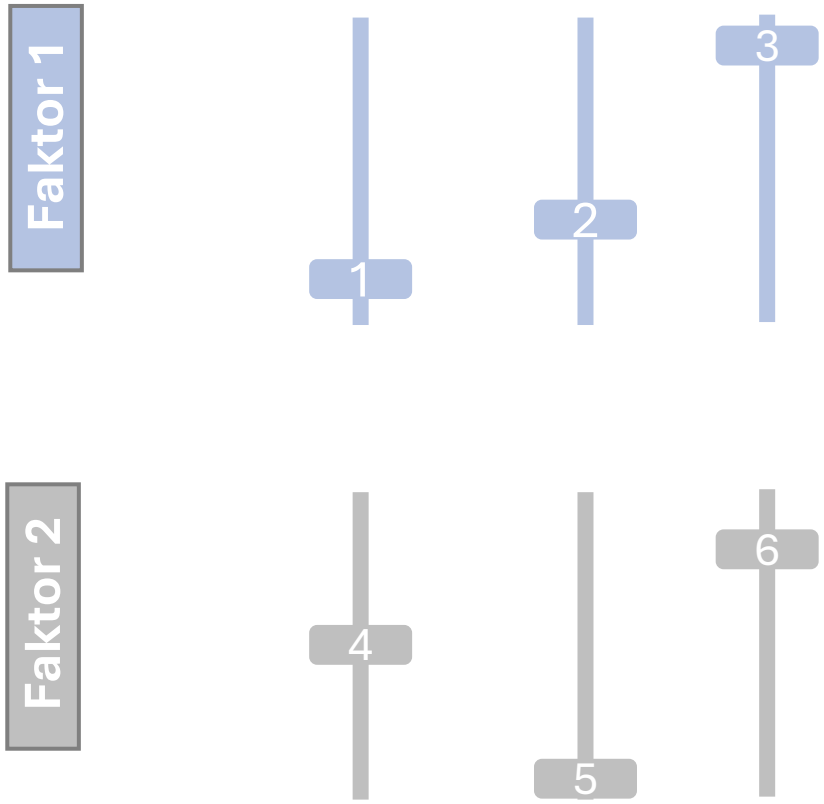


→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

One factor at a time oder Analytical Quality by design?

One Factor at a time



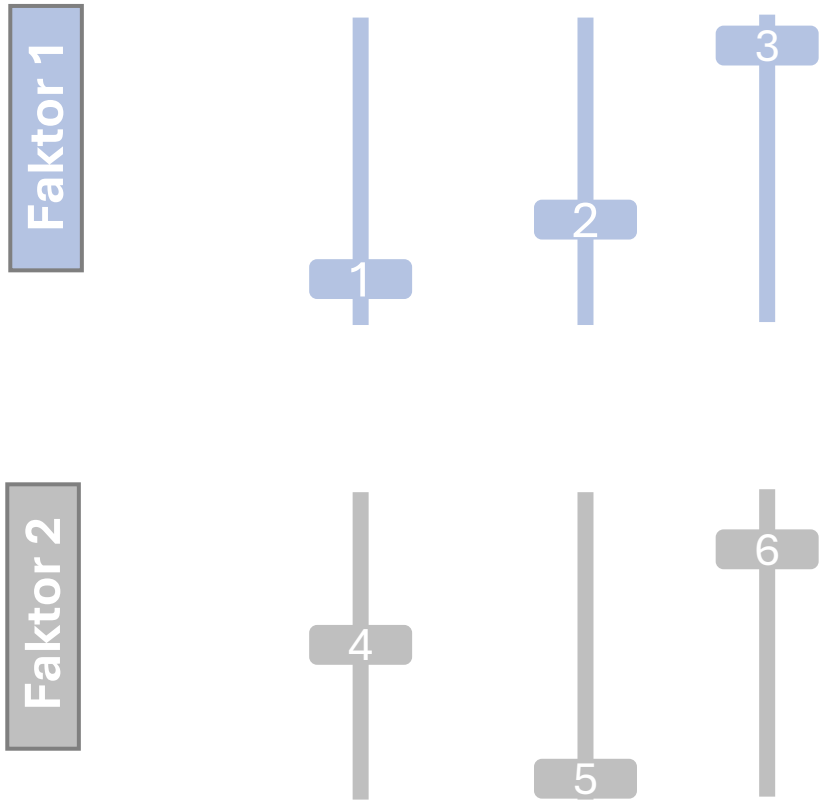
→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

Design of Experiment

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

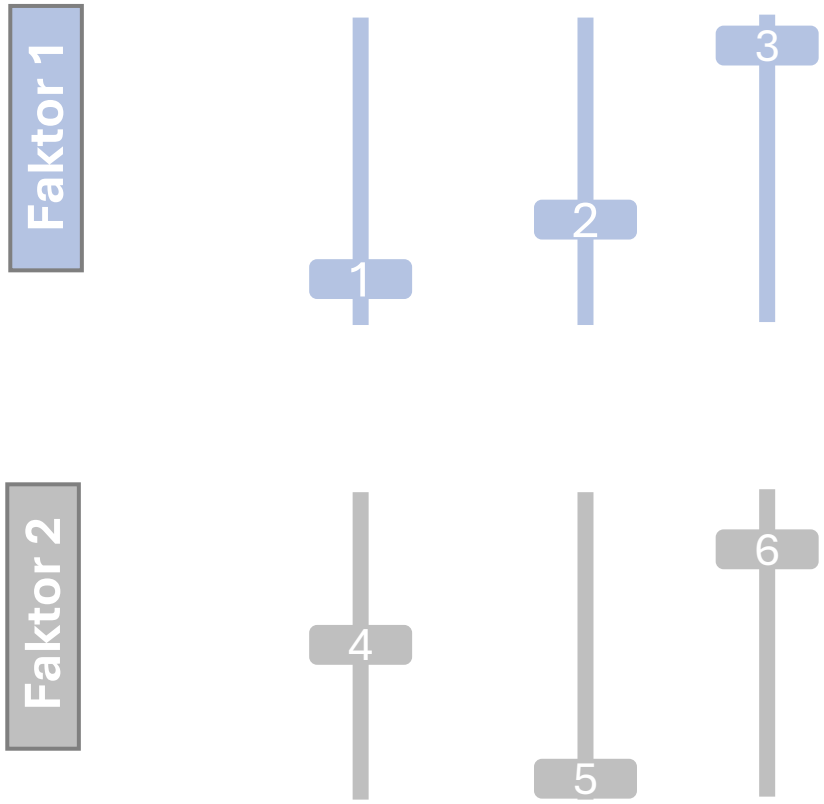
Analytical Quality by Design (AQbD,ICH Q14)

Design of Experiment

Screening

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

Design of Experiment

Screening

Plackett-Burman-Design

5 Faktoren

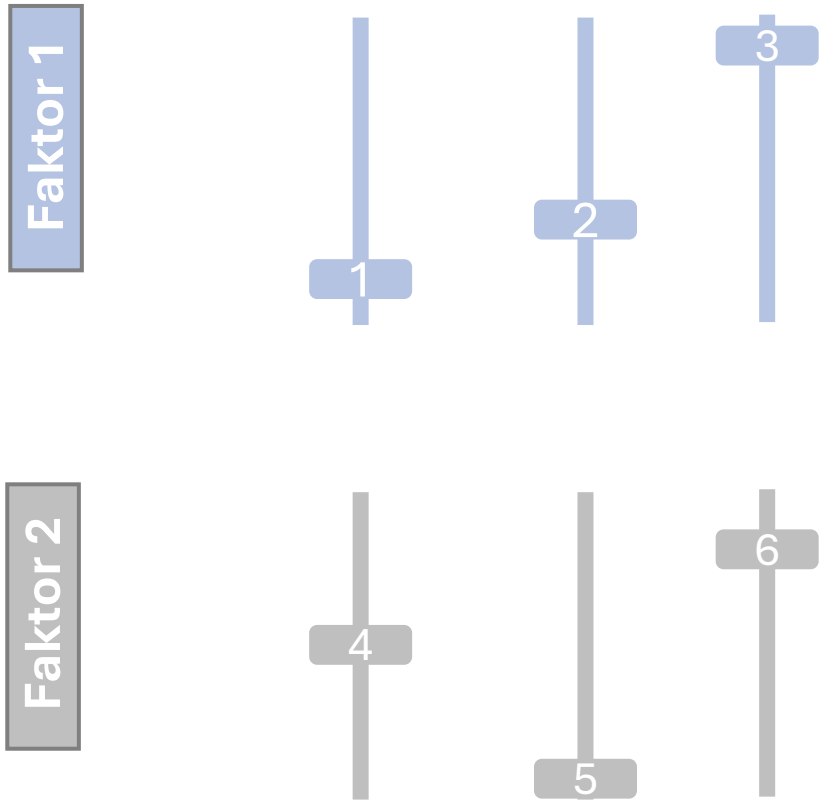
12 Runs

Obidoxim

(1) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q14: Analytical Procedure Development, (2023). (2) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q9: Quality risk management, (2025).
(3) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised guideline Q8(R2): Pharmaceutical development, (2009). (4) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q10: Pharmaceutical quality system, (2008).

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

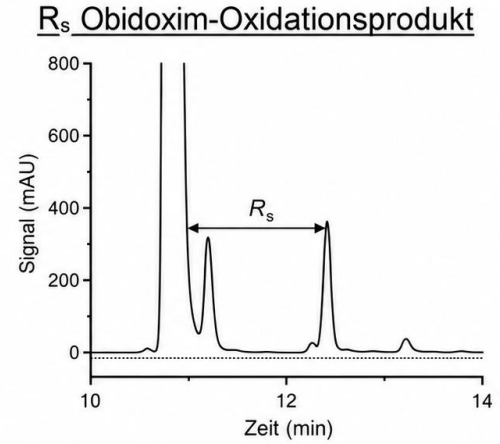
Design of Experiment

Screening

Plackett-Burman-Design

5 Faktoren

12 Runs

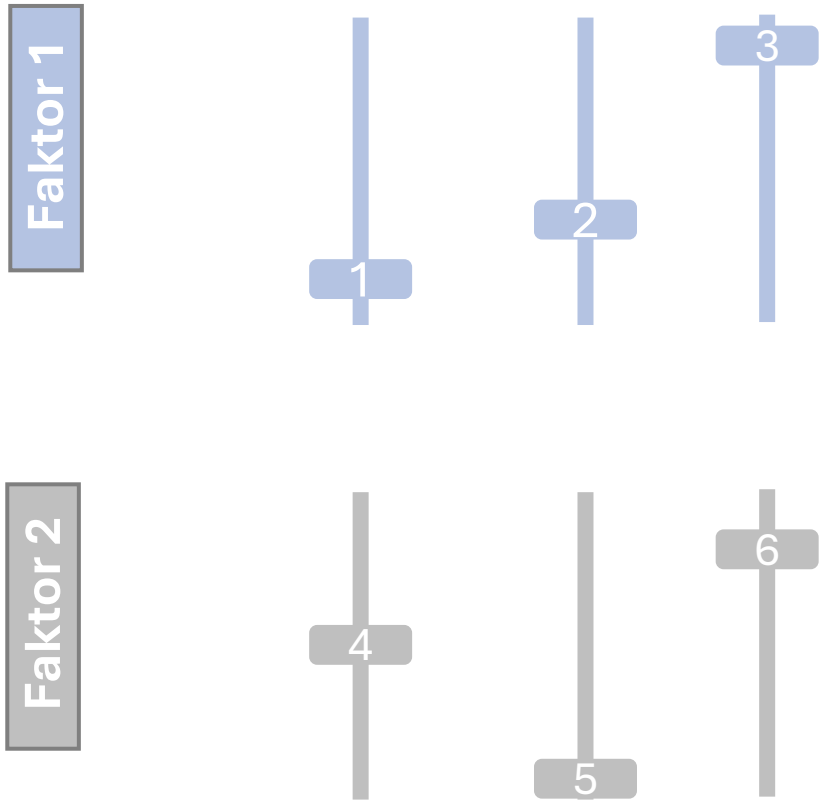


Obidoxim

(1) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q14: Analytical Procedure Development, (2023). (2) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q9: Quality risk management, (2025).
(3) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised guideline Q8(R2): Pharmaceutical development, (2009). (4) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q10: Pharmaceutical quality system, (2008).

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

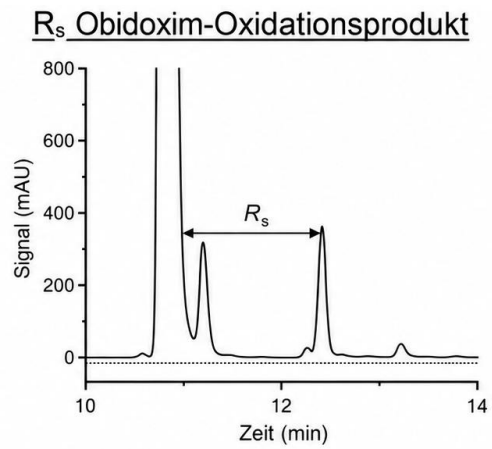
Design of Experiment

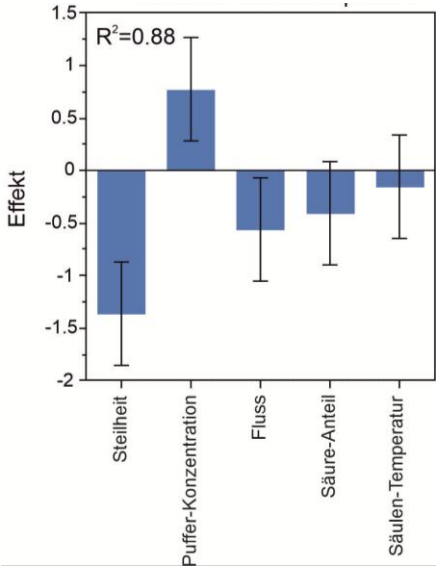
Screening

Plackett-Burman-Design

5 Faktoren

12 Runs



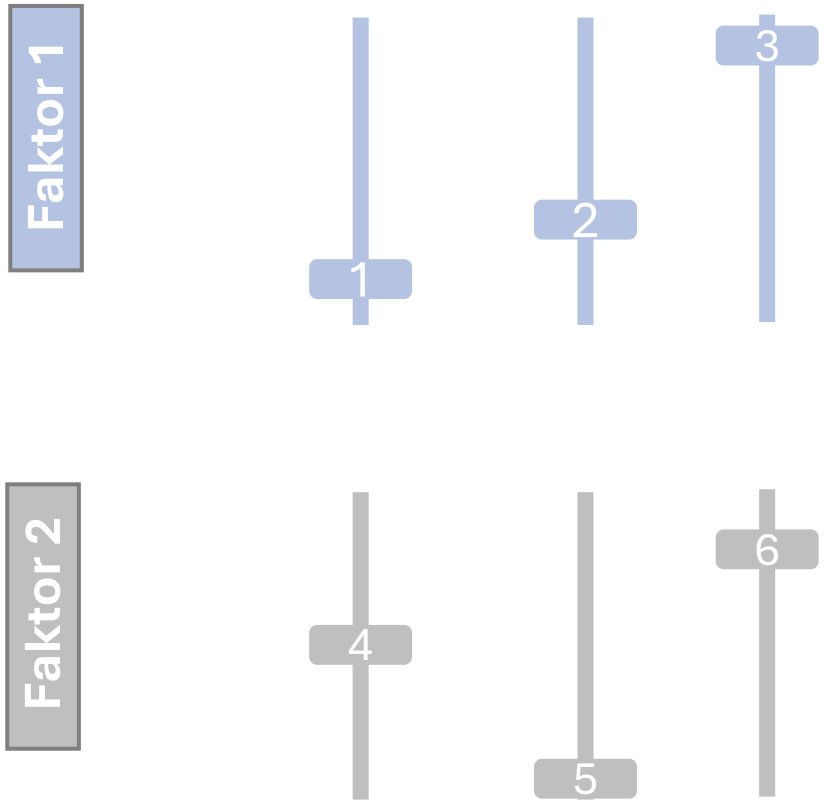


Obidoxim

(1) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q14: Analytical Procedure Development, (2023). (2) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q9: Quality risk management, (2025).
(3) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised guideline Q8(R2): Pharmaceutical development, (2009). (4) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q10: Pharmaceutical quality system, (2008).

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

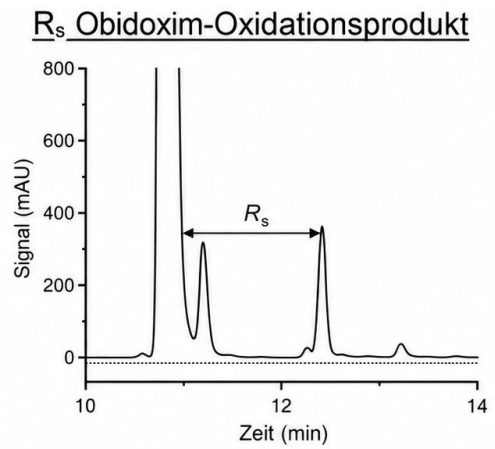
Design of Experiment

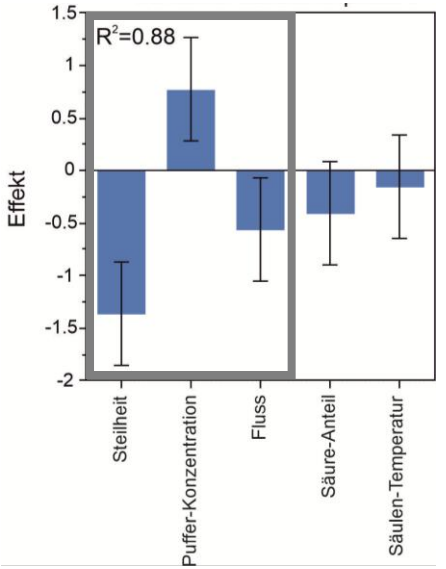
Screening

Plackett-Burman-Design

5 Faktoren

12 Runs



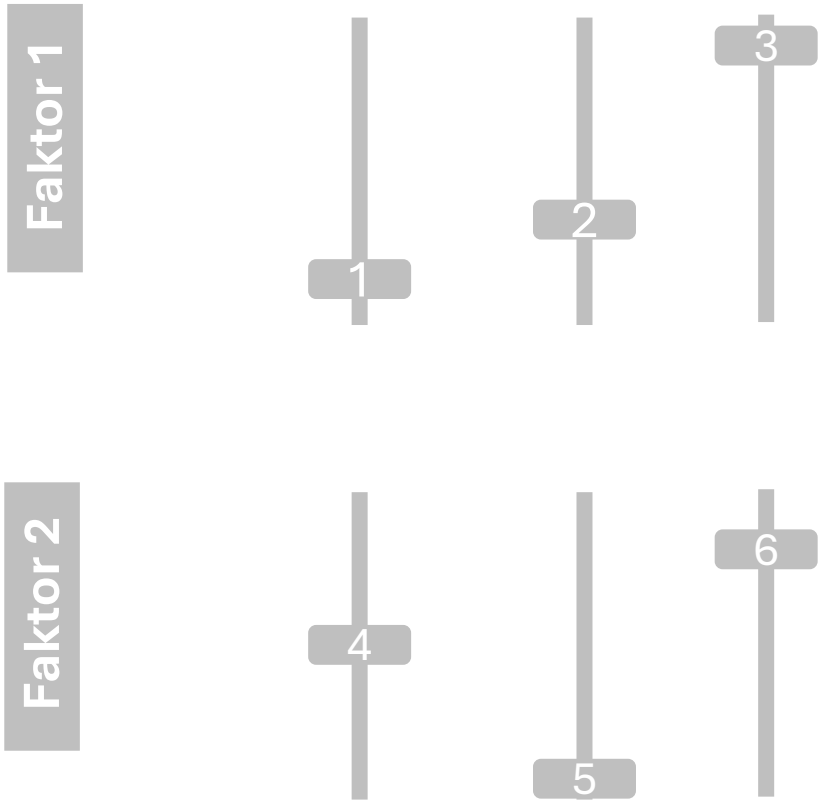


Obidoxim

(1) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q14: Analytical Procedure Development, (2023). (2) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q9: Quality risk management, (2025).
(3) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised guideline Q8(R2): Pharmaceutical development, (2009). (4) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q10: Pharmaceutical quality system, (2008).

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

Design of Experiment

Screening

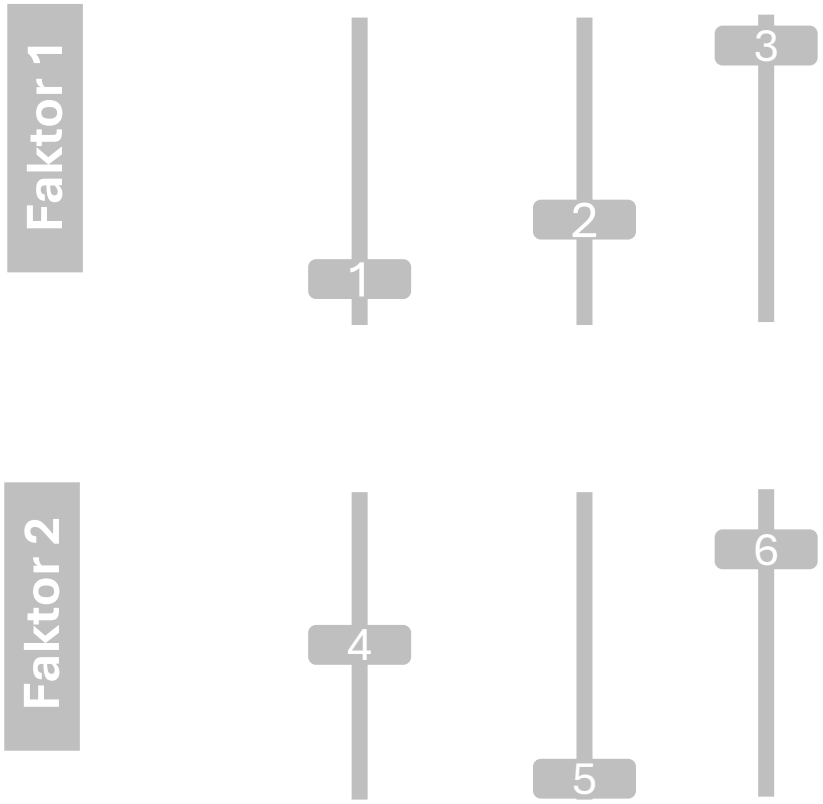
Plackett-Burman-Design
5 Faktoren
12 Runs

Statistisch signifikanter Einfluss von:

+	-
Puffer-Konzentration	Gradienten-Steilheit
	Fluss

One factor at a time oder Analytical Quality by design?

One Factor at a time



→fehlendes Methodenverständnis / Robustheit

Analytical Quality by Design (AQbD,ICH Q14)

Design of Experiment

Screening

Plackett-Burman-Design
5 Faktoren
12 Runs

Statistisch signifikanter Einfluss von:

+	-
Puffer-Konzentration	Gradienten-Steilheit
	Fluss

Optimization

Obidoxim

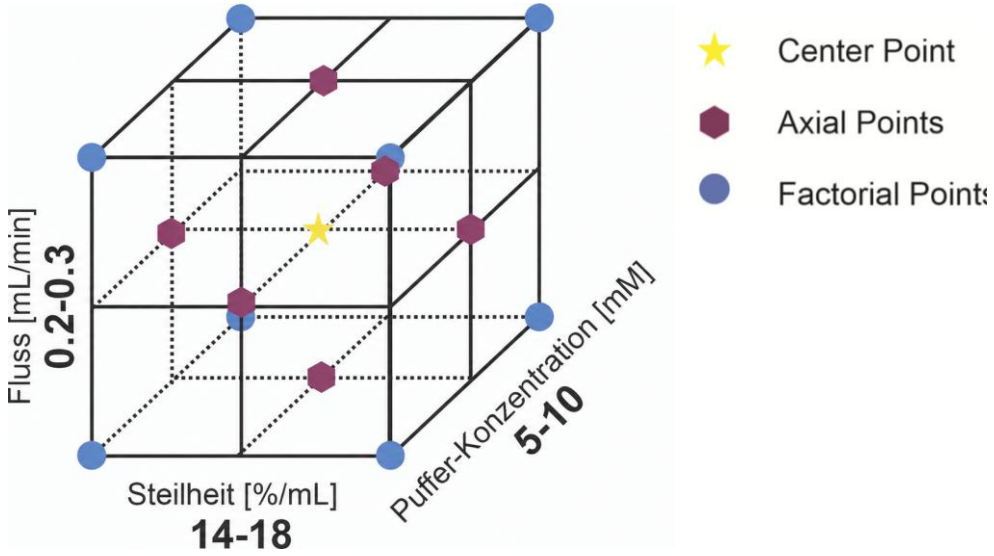
(1) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q14: Analytical Procedure Development, (2023). (2) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q9: Quality risk management, (2025).
(3) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised guideline Q8(R2): Pharmaceutical development, (2009). (4) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use, ICH Harmonised Guideline Q10: Pharmaceutical quality system, (2008).

Optimization: Face Centered Central Composite Design

Design

- Face-Centered Central Composite

→Quadratisches Modell **mit** Interaktionen



Obidoxim

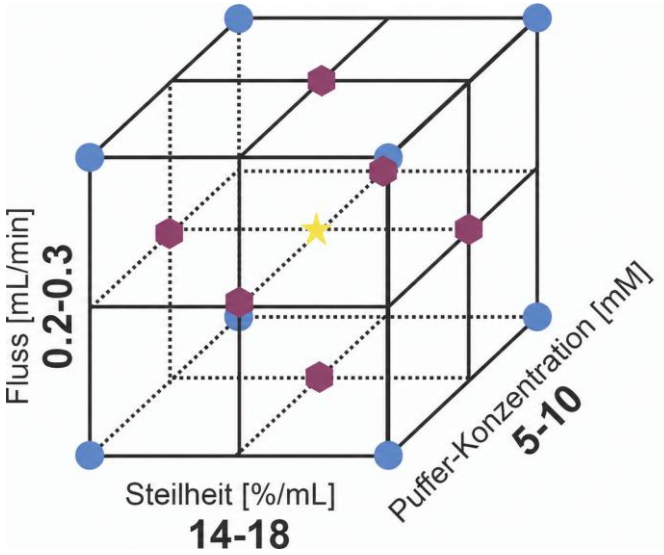
(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

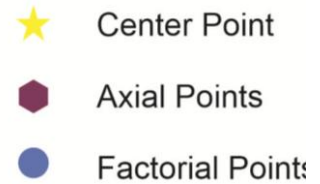
Optimization: Face Centered Central Composite Design

Design

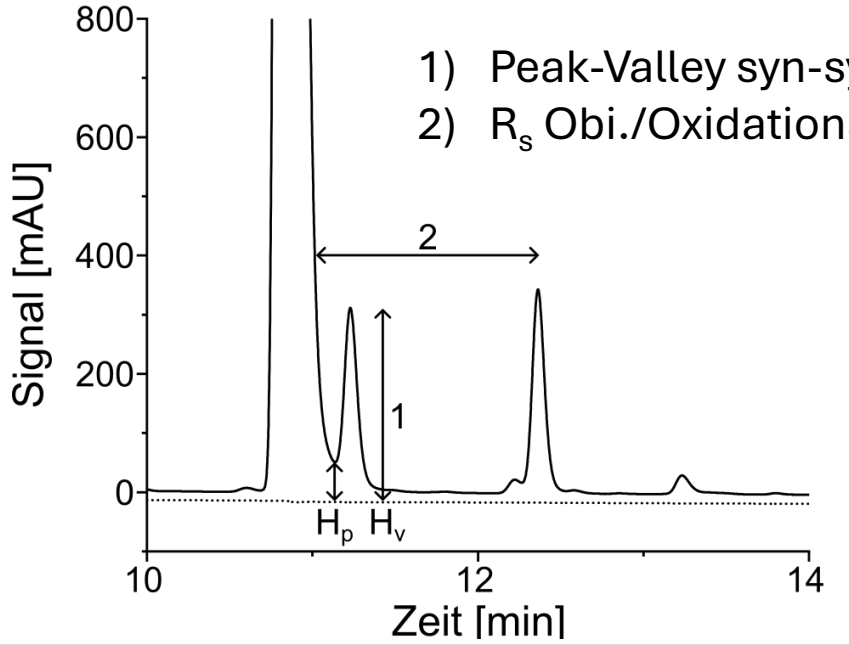
- Face-Centered Central Composite

→Quadratisches Modell **mit** Interaktionen





Responses



1) Peak-Valley syn-syn/syn-anti
2) R_s Obi./Oxidationsprodukt

Obidoxim

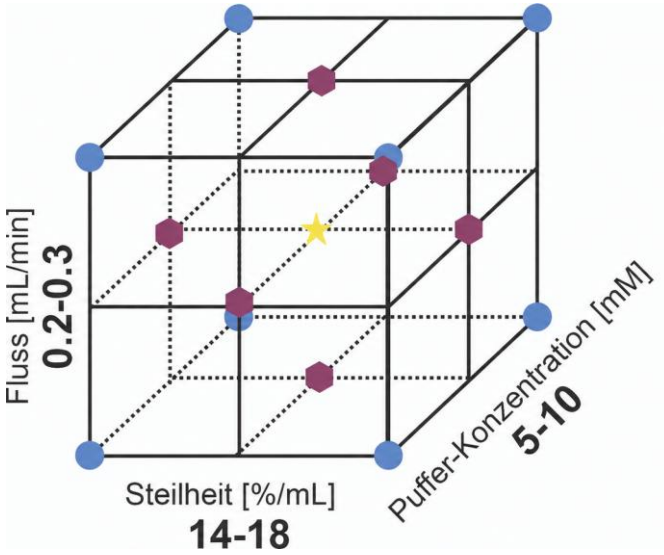
(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

Optimization: Face Centered Central Composite Design

Design

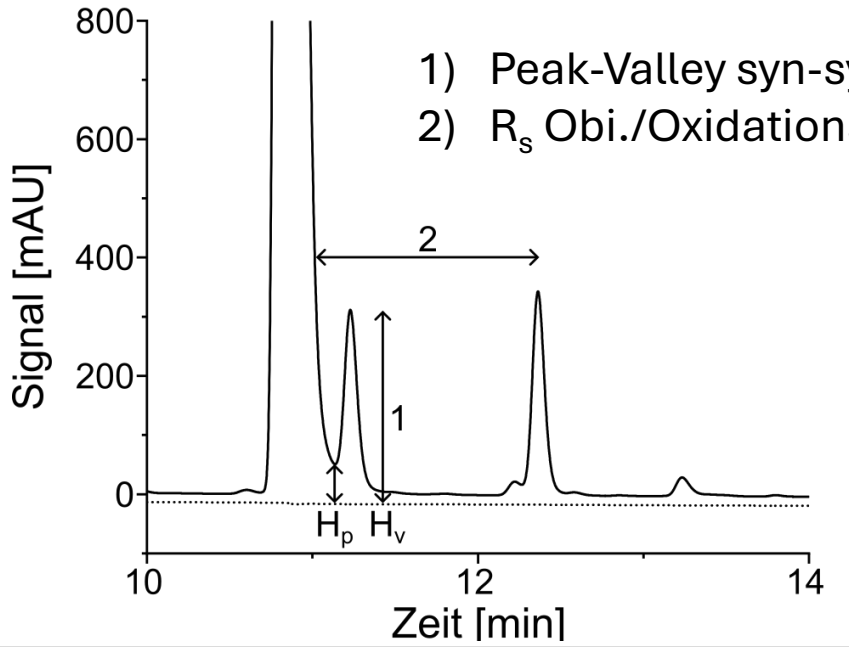
- Face-Centered Central Composite

→Quadratisches Modell **mit** Interaktionen

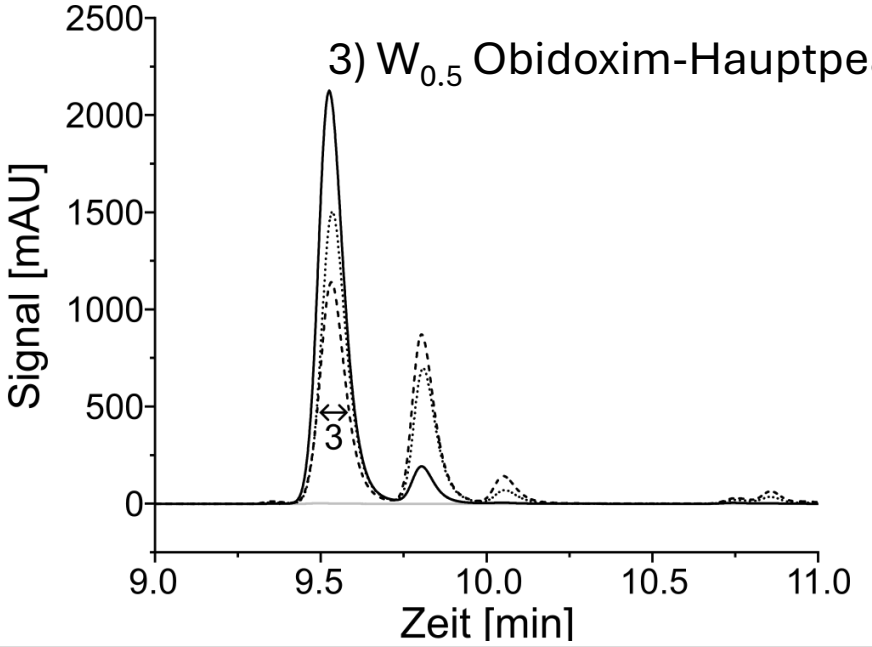


Responses

1) Peak-Valley syn-syn/syn-anti
2) R_s Obi./Oxidationsprodukt



3) $W_{0.5}$ Obidoxim-Hauptpeak im Assay



Obidoxim

(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

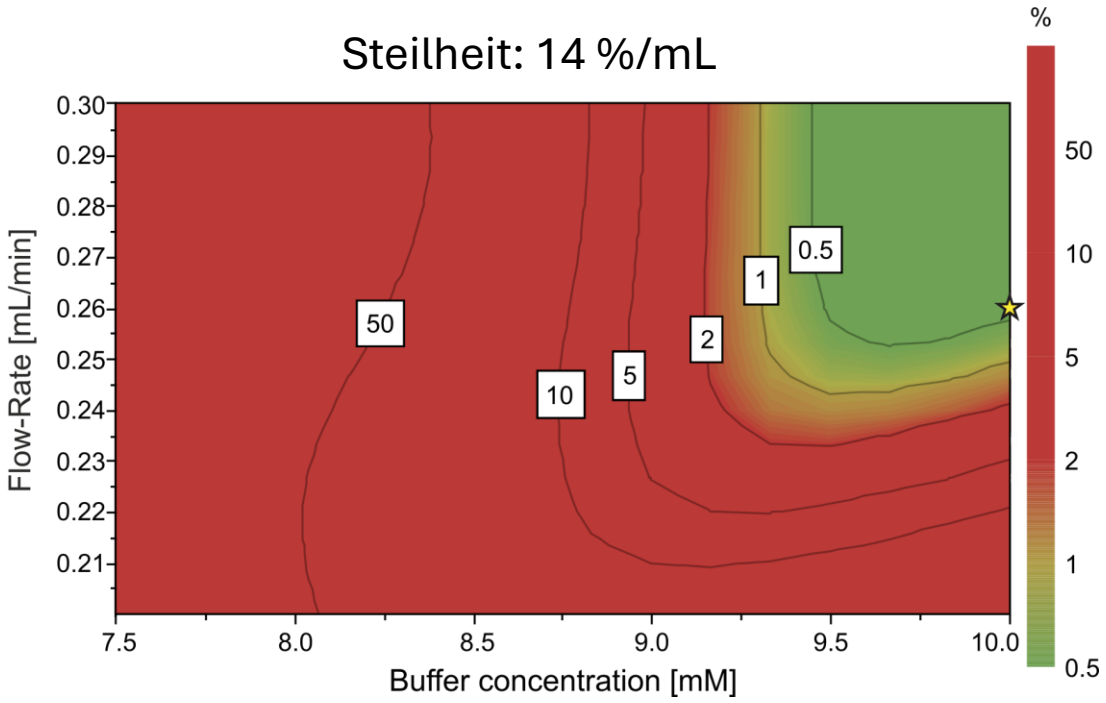
Optimization: Auswertung

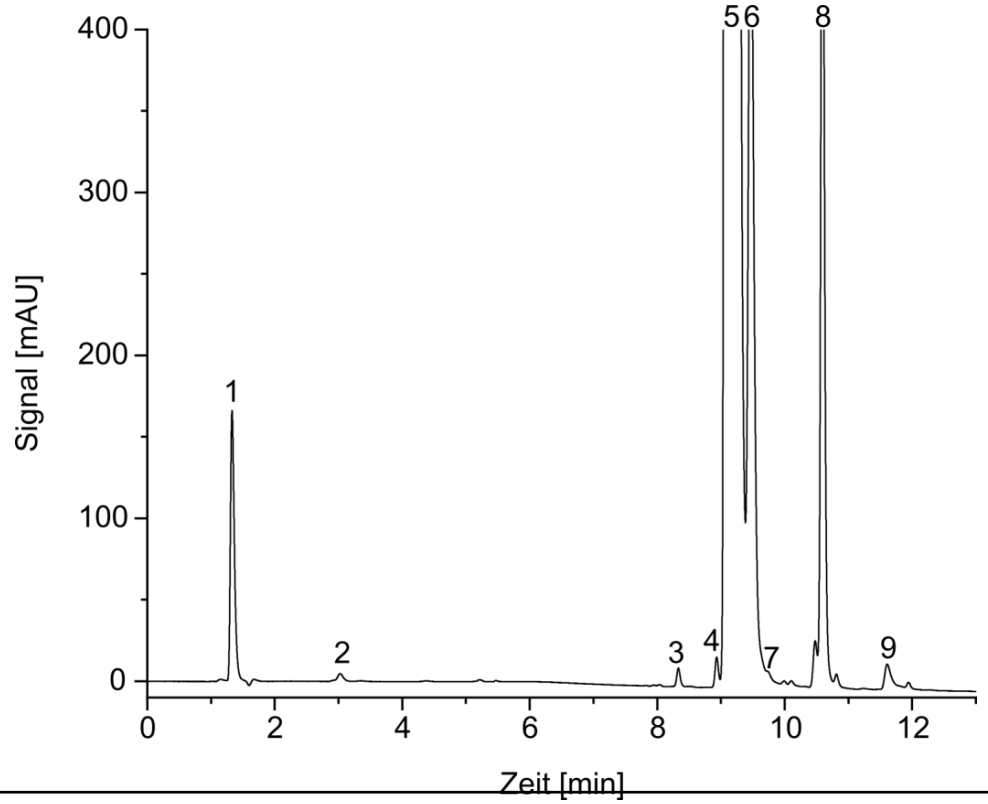
Vorgehen

1. Durchführen einer Regressionanalyse
2. ANOVA

→Entfernen der **nicht-signifikanten** Terme (p > 0.05)

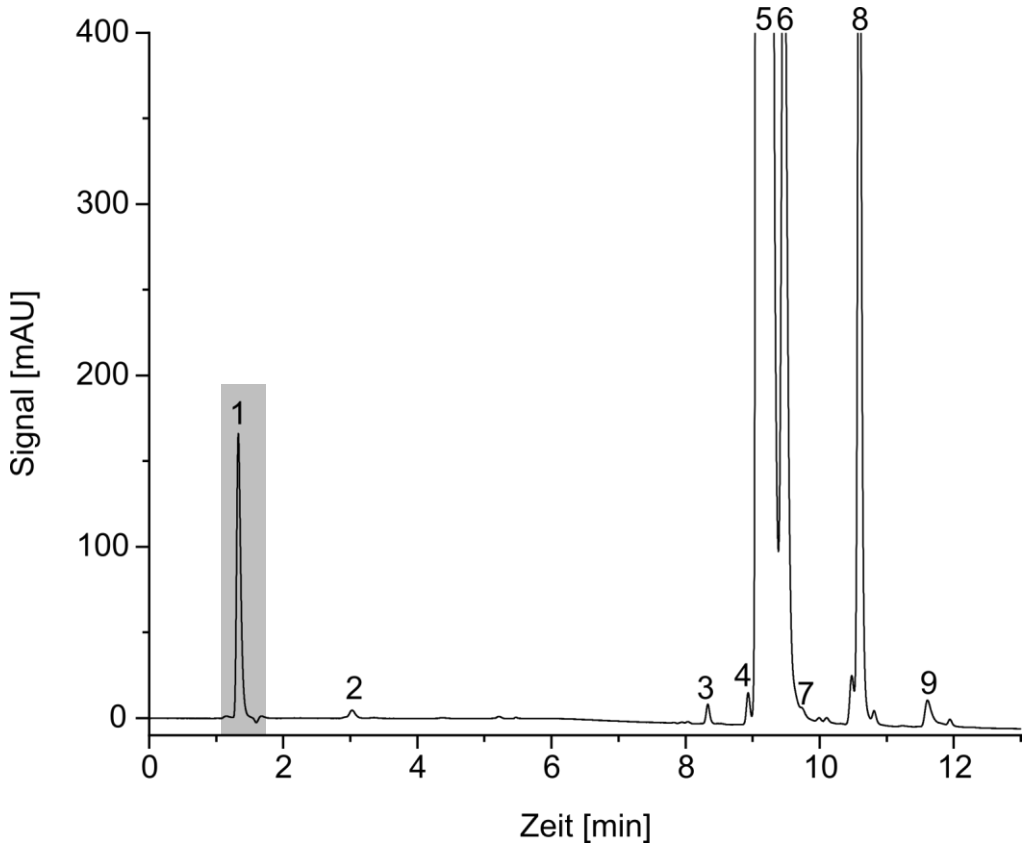
3. Festlegen der Akzeptanzkriterien für Responses
4. Monte-Carlo-Simulationen (Finden der Method Operable Design Region)

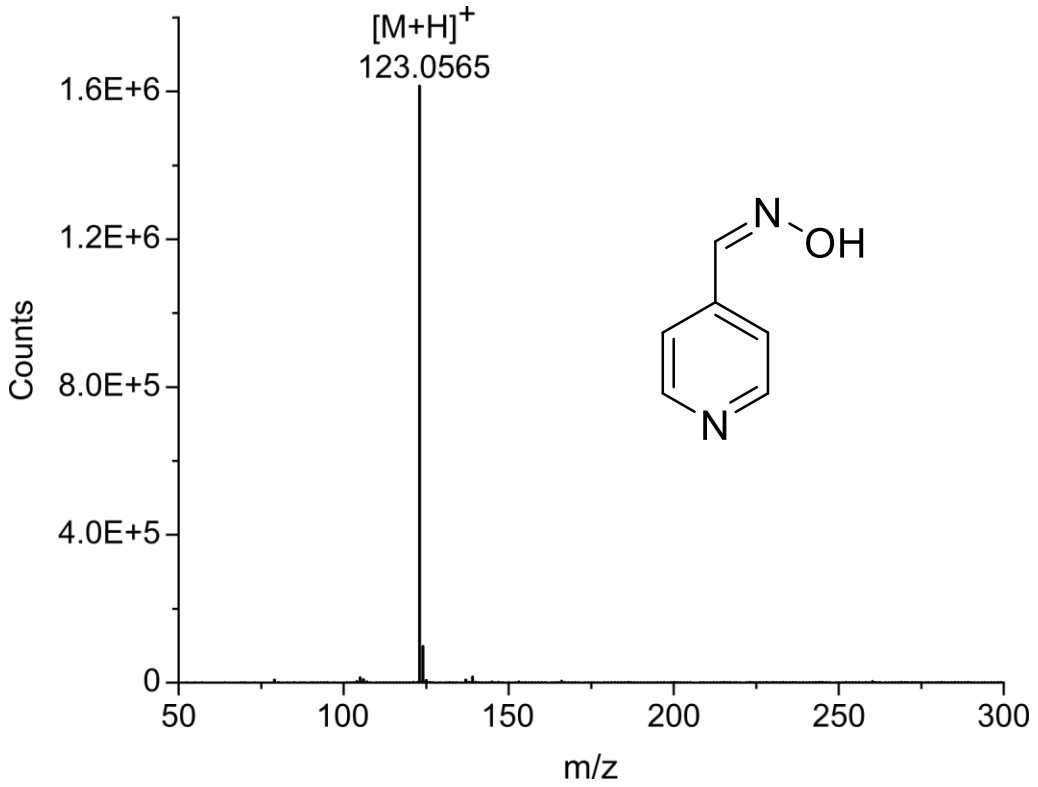




(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

QToF: Peak 1



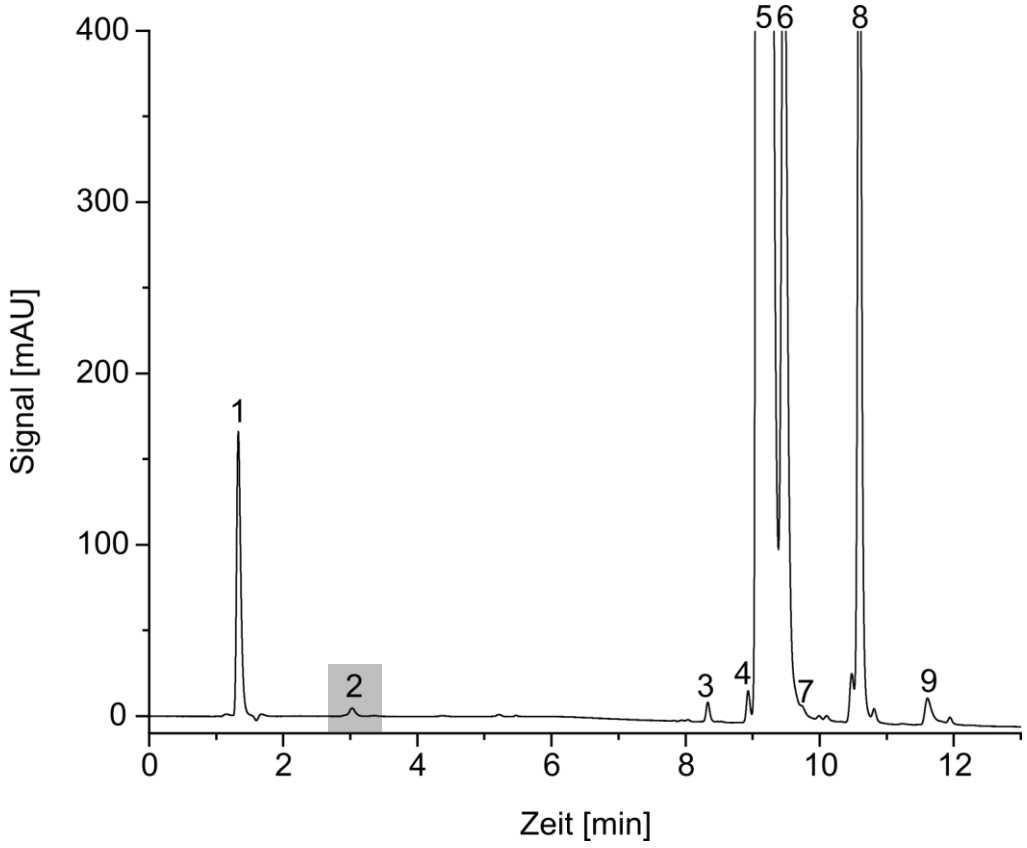


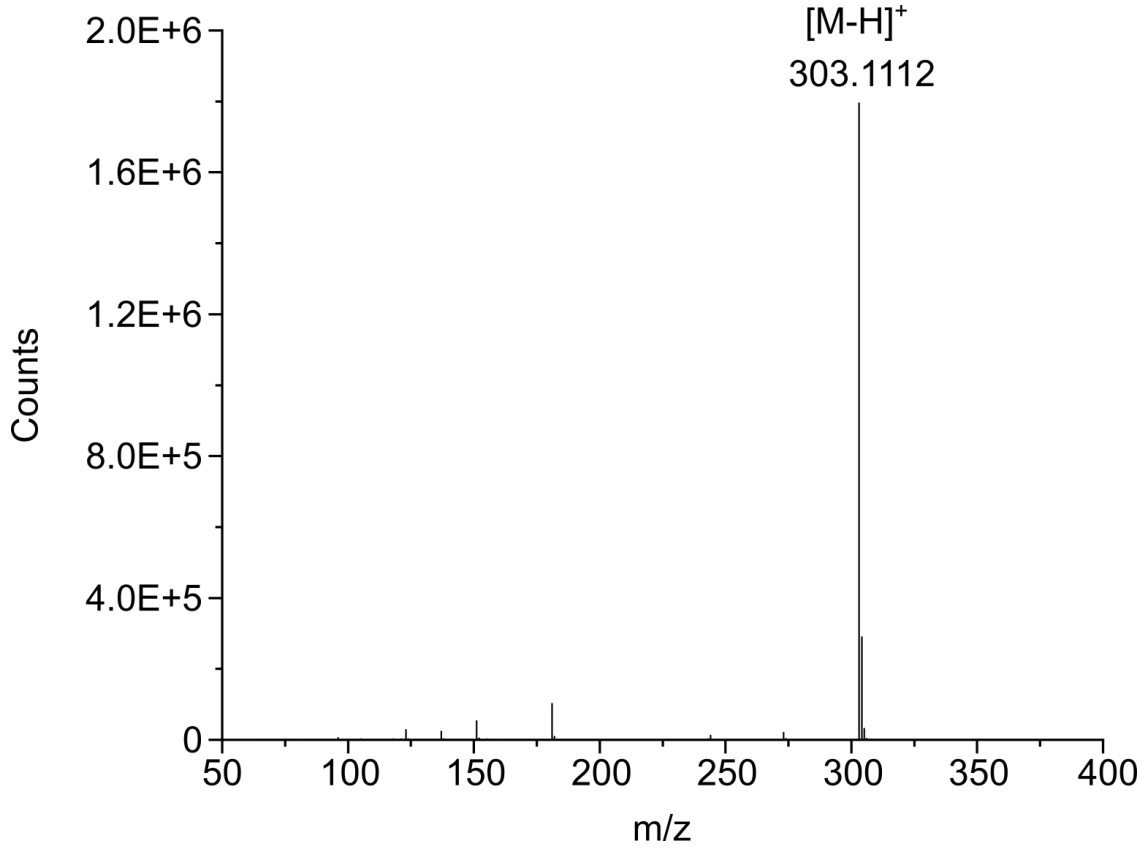
	Masse rechnerisch	Masse gemessen	Unterschied [ppm]
$[M+H]^+$	123.0553	123.0565	+ 9.8

Obidoxim

(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

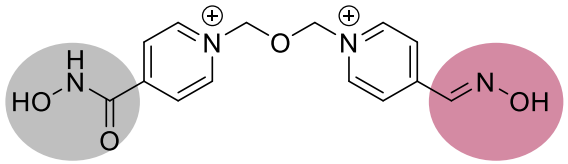
QToF: Peak 2



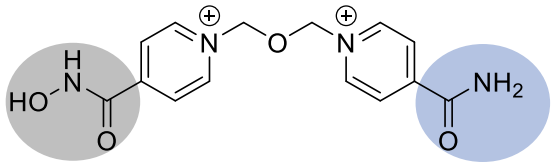


(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

QToF: Peak 2



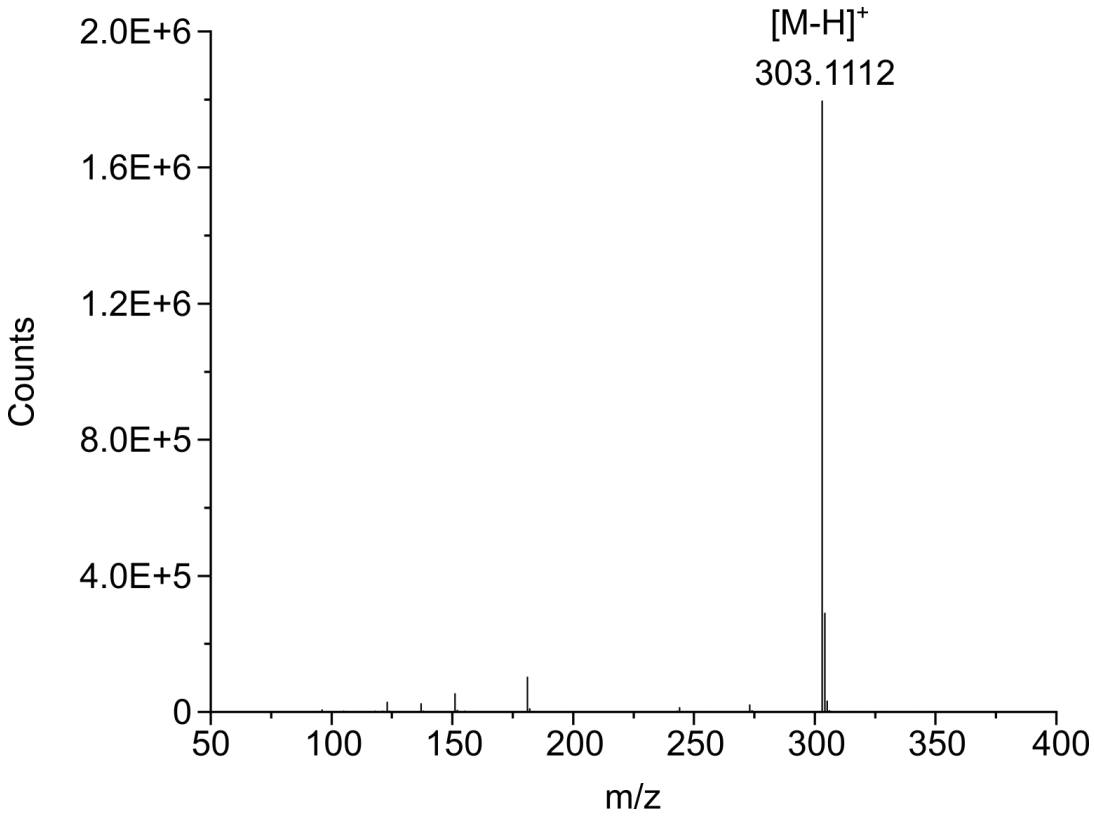
Hydroxamsäure + Oxim



Hydroxamsäure + Amid

→ **Exakt gleiche Massen beider Strukturen!**

	Masse rechnerisch	**Masse gemessen**	**Unterschied [ppm]**
$[M-H]^+$	303.1088	303.1112	+ 7.9
$[ISF]^+$	181.0608	181.0620	+ 6.6



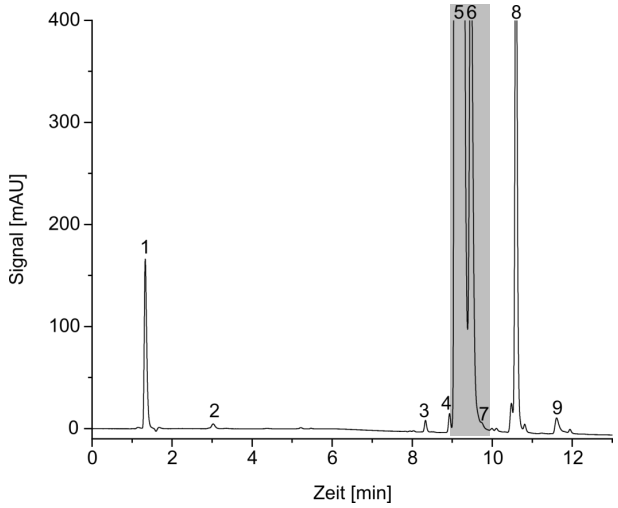
ISF: In-Source-Fragment

Obidoxim

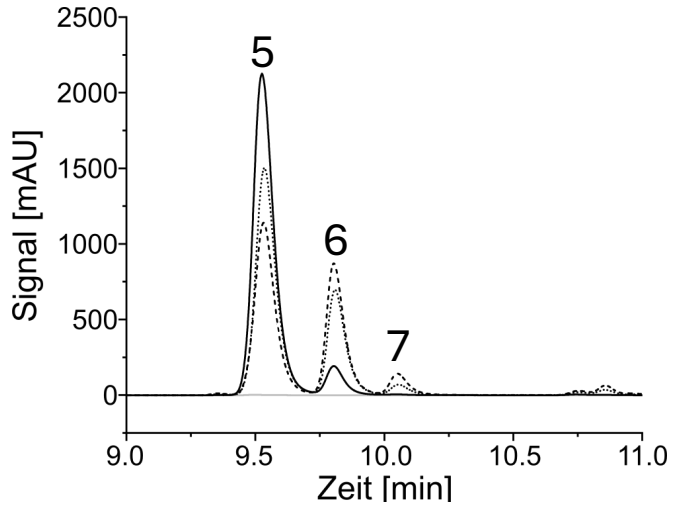
(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript. (2) V. Voicu, F. Albu, F. Tache, K. Musilek, K. Kuca, A. Medvedovici, LC–MS/MS approaches for the assay of bis-quaternary pyridinium oximes used as AChE reactivators in biological matrices, Bioanalysis, 5 (2013) 793–809.

QToF: Obidoxim-Isomere

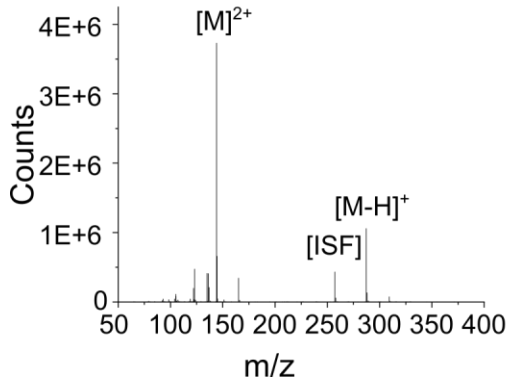
ATOX II 10 mg/mL



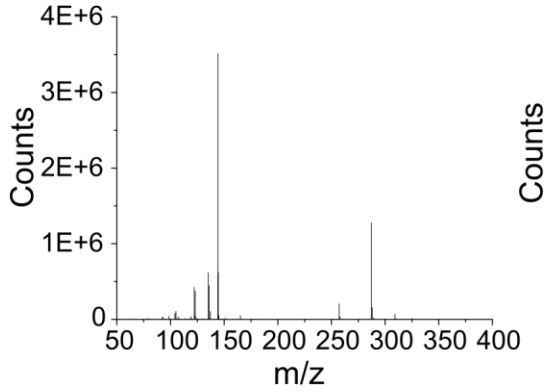
UV-Degradation (0.5 mg/mL)



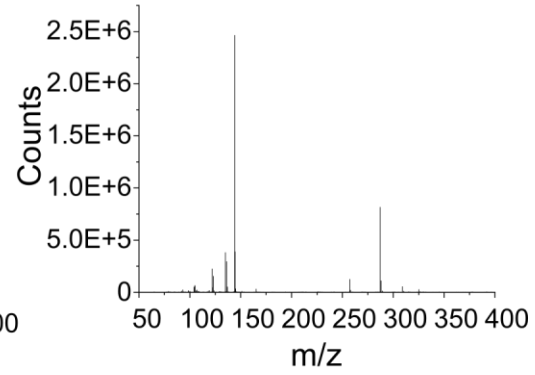
Peak 5



Peak 6



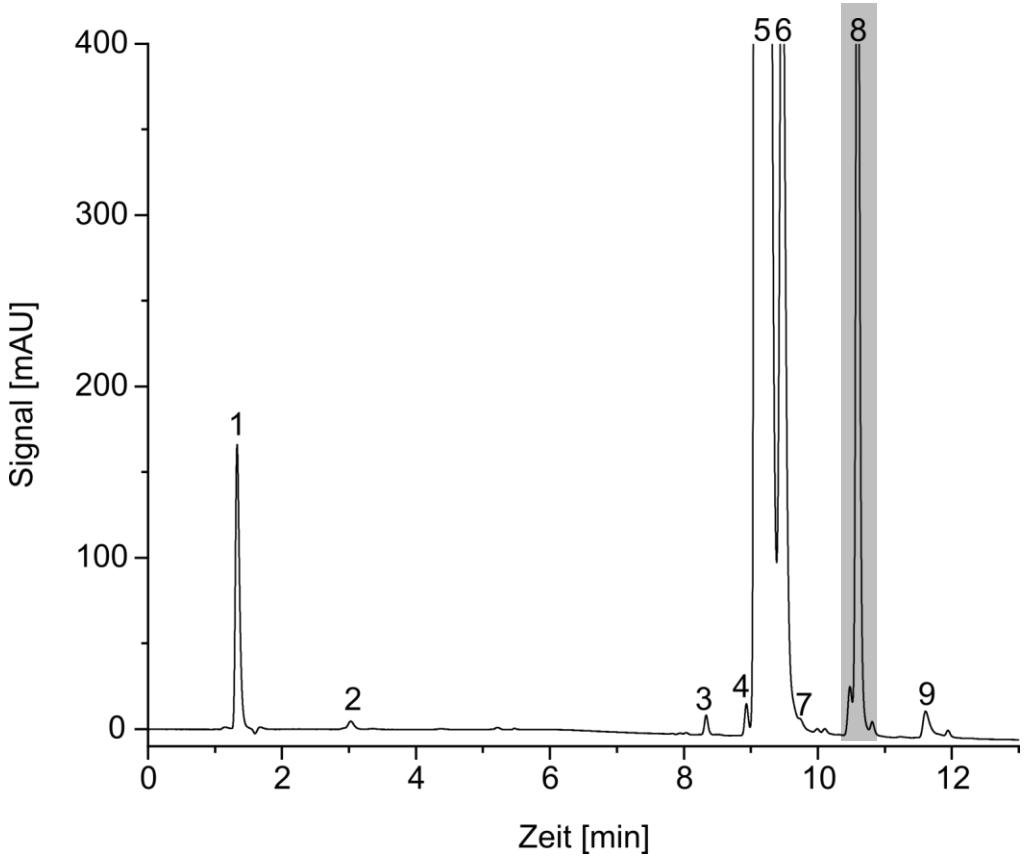
Peak 7

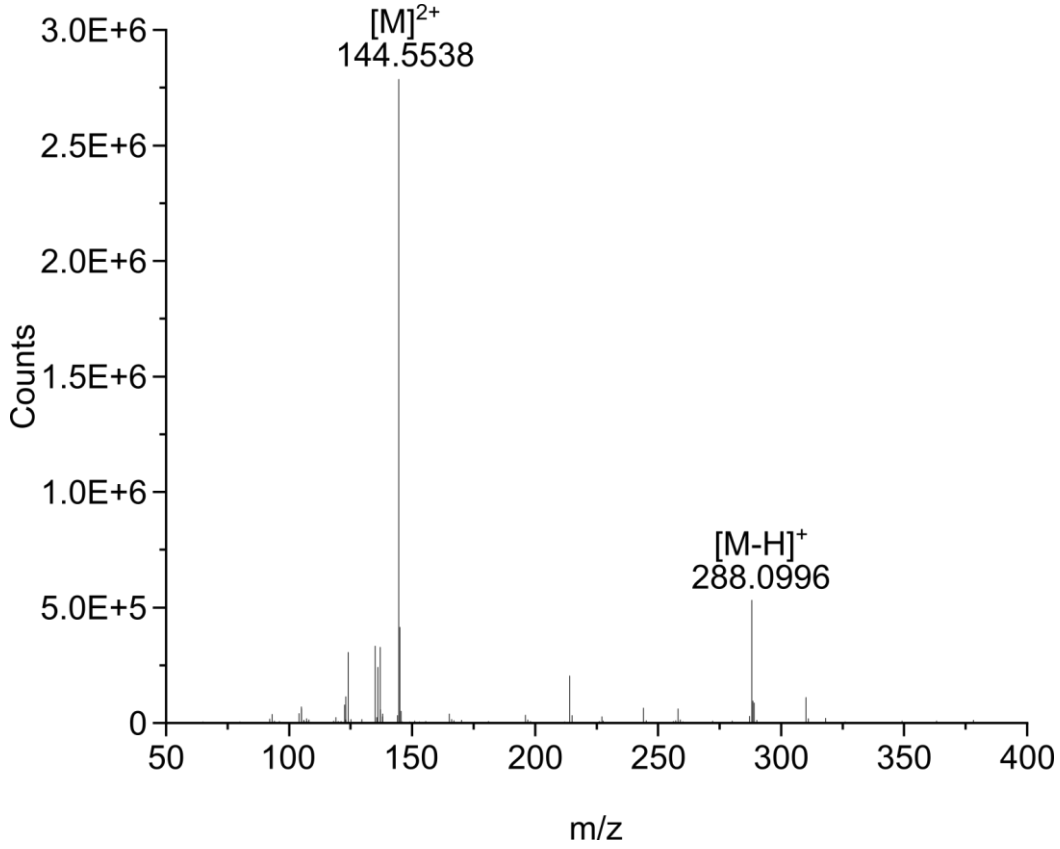


Obidoxim

(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

QToF: Peak 8

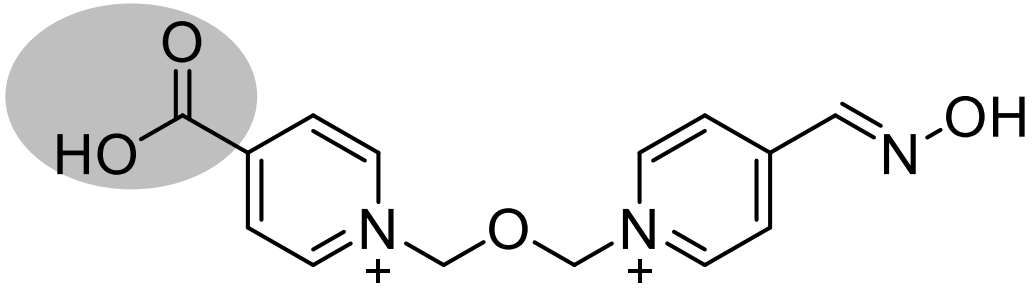




Obidoxim

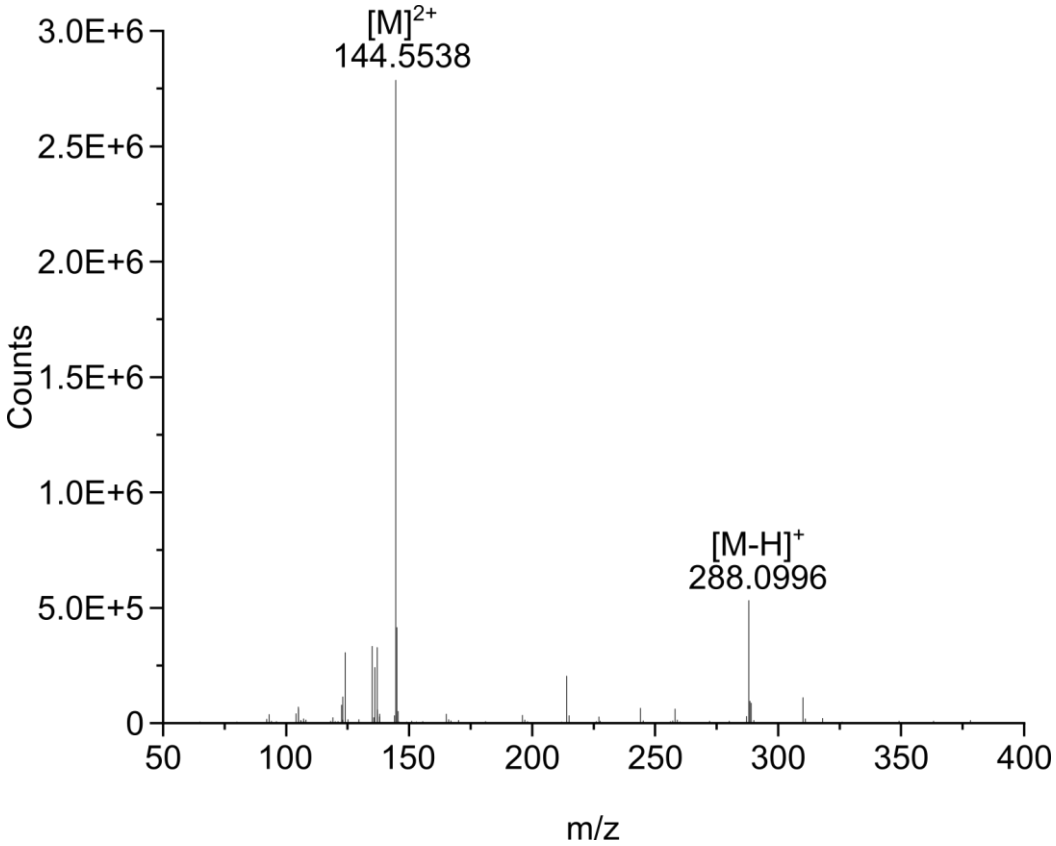
(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript.

QToF: Peak 8



Obidoxim-Monocarbonsäure

	Masse rechnerisch	Masse gemessen	Unterschied [ppm]
$[M-H]^+$	288.0990	288.0996	+ 2.08
$[M]^+$	144.5531	144.5538	+ 4.84



(1) Sperber et al., Impurity profiling of ready-to-use obidoxime autoinjectors: An Analytical Quality by design (AQbD) driven HILIC method development and pharmacological consequences of photoisomerism, unpublished manuscript. (2) V. Voicu, F. Albu, F. Tache, K. Musilek, K. Kuca, A. Medvedovici, LC–MS/MS approaches for the assay of bis-quaternary pyridinium oximes used as AChE reactivators in biological matrices, Bioanalysis, 5 (2013) 793–809.

Fazit und Ausblick

- ✅ Erfolgreiche Methodenentwicklung im AQbD-Konzept
- ❗ Identifizierung von Hauptverunreinigungen

  → Keine eindeutige Zuordnung nur mit LC-HRMS oder LC-MS/MS

  - ➢ Synthese der vermuteten Verunreinigungen
  - ➢ NMR-Messungen der Verunreinigungs-Peaks

Ausblick

Danke für ihre Aufmerksamkeit!

Ich freue mich auf ihre Fragen!

