

Development of ISO Standards for the determination of ammonia, nitrate and total nitrogen using tube tests



Water Analysis

Dr. Christian Prokisch, 26.07.2022

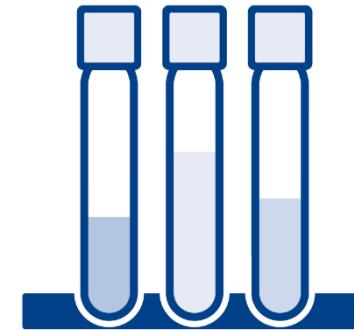
Agenda



Project idea



Companies



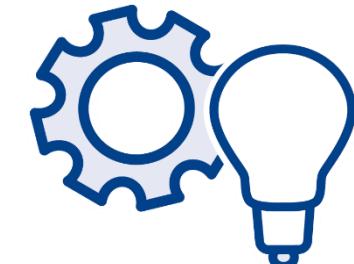
Components



Interlaboratory
trial design



Ad Block



Summary



Project idea

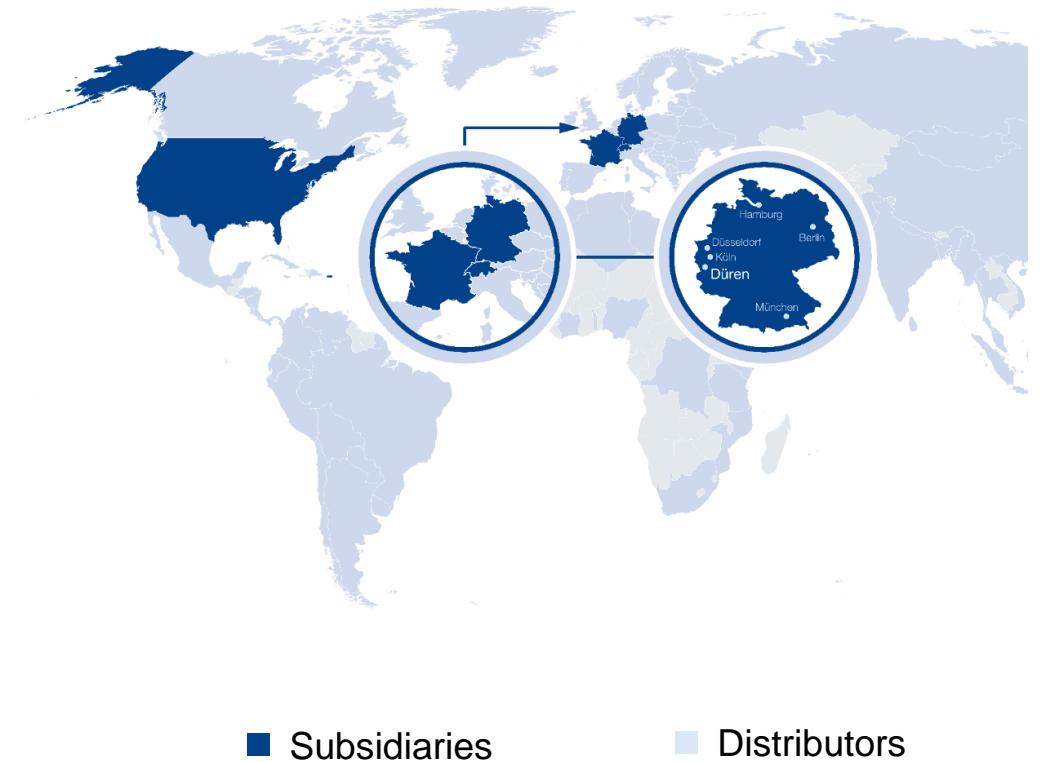




Project idea

MACHEREY-NAGEL Basic figures

- 4th Generation family owned
- More than 700 employees
- More than 25.000 products
- Turnover 150 Mio. €

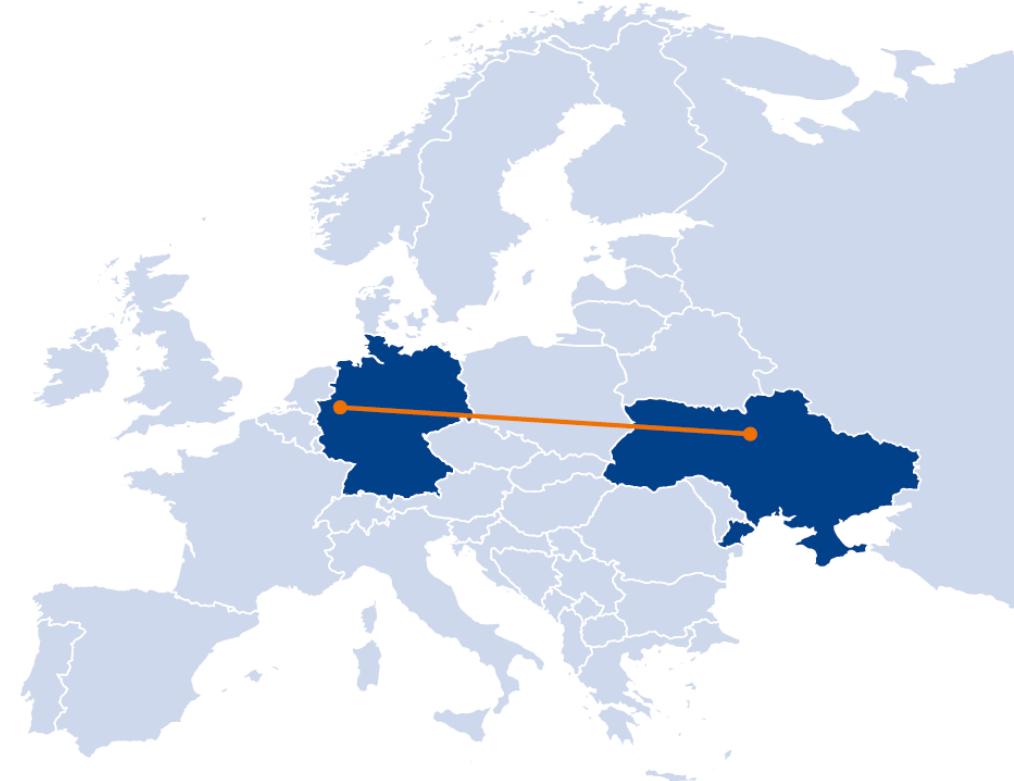




Project idea

No war related supply chain issues

- Energy consumption comparably small
- Very high production depth in –house
- Comparably small supply chain issues
- Just for information:
 - Distance Düren – Kiew = 2,000 km or 21 hours (Google)





ISO Projects





ISO Projects

Small scale sealed tubes

- Ammonium
 - ISO DIS 23695
- Nitrate
 - ISO DIS 23696 – 1: Dimethylphenol
 - ISO DIS 23696 – 2: Chromotropic Acid
- Total Nitrogen
 - ISO DIS 23697 – 1: Dimethylphenol
 - ISO DIS 23697 – 2: Chromotropic Acid





ISO Projects

The way

- Project suggested to WG81 by Italy
- First discussed in Tokio, 2019
 - Hurricane affected meeting
- Resolution:
 - Total Nitrogen Parameter name: ST-TNb
 - Became a huge issue in the following discussion





ISO Projects

The way

- Manufacturer support needed
- The following volunteered:
 - HACH (suggested project)
 - MERCK
 - Lovibond
 - MACHEREY-NAGEL
- All in Germany
- Most of the work was expected of the German mirror group



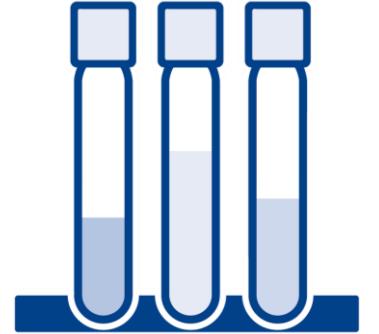


ISO Projects

The way

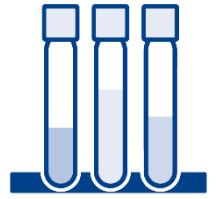
- Christian Prokisch lead this group starting 2020
- Projects got people emotionally involved
 - Online meetings made participation easy
 - Intense discussions in WG 81
 - >1 h discussion at SC2 meeting
- Intense experience





General objections





General objections

Why write an ISO Norm

- Why not?
 - Project was accepted by majority of countries
 - Users appreciate tests according to norm
 - We show that different manufacturers provide similar results
 - Work is mostly done by manufacturers
- Easy tests empower little trained users
- International ILT proves reliability



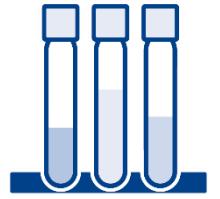


General objections

Proprietary chemistry should not go into norm

- Chemistry is not proprietary
 - Long described in literature
 - Norm discloses substances and concentration ranges
- Many other examples (Colilert, Chlorine)





General objections

ISO must not support a company

- It does not support just 1 company
 - 4 major tube tests manufacturers contribute
- Can it go any worse then IDEXX and Colilert?
- In contrast to ISO We give the analytes + a usable composition range
- Anyone can produce it





Interlaboratory trial design

ISO 23695 „ammonium Nitrogen“



ISO 23696-1 / -2 „nitrate“

ISO 23697-1 / -2 „total nitrogen“



Interlaboratory trial design

ISO must not support a company

- Organizer: AK 30, Christian Prokisch
- Service provider: AQS Baden Würtemberg, Michael Koch
- Statistics: Technical University Hessen, Gerhild Donnevert
- Shipment of samples: February 2022
- Plausibility check of results: April 2022
- Results: May 2022



Interlaboratory trial design

Samples

| Sample ID | Matrix | NO ₃ -N | NH ₄ -N | TNb |
|-----------|---------------------|--------------------|--------------------|------|
| A | Ground water | 1 | 0.1 | n.a. |
| B | Swimming pool water | 7–8 | 0.5 | n.a. |
| C | Waste water 1 | 40 | 3 | n.a. |
| D | Waste water 2 | 8 | 40 | n.a. |
| E | Waste water 3 | n.a. | n.a. | 10 |
| F | Waste water 4 | n.a. | n.a. | 60 |
| G | Waste water 5 | n.a. | n.a. | 80 |
| H | Surface water | 10–12 | 0.8 | n.a. |
| I | QC standard | 1 | 8 | 13 ← |

For TNb Glycin was used for spiking => interference for Ammonium test described in DIS



ISO 23695 “ammonium nitrogen”





ISO 23695 “ammonium nitrogen”

Composition information

Salicylate reaction

| Reagent name | Mass fraction range (% w/w) |
|-----------------------------|--------------------------------|
| Sodium dichloroisocyanurate | 0.004 to 0.05 |
| Sodium salicylate | 0.1 to 5 |
| Sodium nitroprusside | 0.01 to 5 |

Phenol-substitute based reaction

| Reagent name | Mass fraction range (% w/w) |
|-----------------------------|--------------------------------|
| Sodium dichloroisocyanurate | 0.008 to 0.05 |
| Chlorophenol | 0.01 to 0.1 |
| Sodium nitroprusside | 0.05 to 0.5 |



ISO 23695 “ammonium nitrogen”

Procedure

NANOCOLOR® Ammonium 3
Amonio

Methode / Method / Méthode / Método 690 nm Test 0-03
0031 0.04–2.30 mg/L NH₄-N Min 20–80 % Max
0032 0.05–3.00 mg/L NH₄⁺ 0.04 0.49 1.85 2.30 mg/L NH₄-N
0033 0.05–3.00 mg/L NH₃ 0.05 0.64 2.41 3.00 mg/L NH₄⁺/NH₃

1 4.0 mL pH 7–10 2 1 x R2 3 4 15 min 5

6 M

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ISO 23695 “ammonium nitrogen”

Symbols used in results tables

| | |
|-----------|---|
| I | Number of laboratories after outlier rejection |
| n | Number of individual test results after outlier rejection |
| o | Percentage of outliers |
| X | Assigned value |
| \bar{x} | Overall mean of results (without outliers) |
| η | Recovery rate |
| s_R | Reproducibility standard deviation |
| $C_{V,R}$ | Coefficient of variation of reproducibility |
| s_r | Repeatability standard deviation |
| $C_{V,r}$ | Coefficient of variation of repeatability |



ISO 23695 “ammonium nitrogen”

ILT results

| Sample | Matrix | I | n | o | X | \bar{x} | η | s_R | $C_{V,R}$ | s_r | $C_{V,r}$ |
|--------|---------------------|----|-----|------|-----------------|-----------------|--------|-----------------|-----------|-----------------|-----------|
| | | | | % | $\mu\text{g/L}$ | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % |
| A | Ground water | 38 | 152 | 17,4 | - | 0,145 | - | 0,013 | 9,0 | 0,004 | 2,8 |
| B | Swimming pool water | 43 | 171 | 9,0 | - | 0,845 | - | 0,045 | 5,3 | 0,014 | 1,7 |
| C | Waste water 1 | 39 | 155 | 17,6 | - | 4,192 | - | 0,211 | 5,0 | 0,055 | 1,3 |
| D | Waste water 2 | 46 | 184 | 4,2 | - | 45,013 | - | 2,375 | 5,3 | 0,867 | 1,9 |
| H | Surface water | 41 | 164 | 10,9 | - | 0,789 | - | 0,033 | 4,2 | 0,012 | 1,5 |
| I | QC Standard | 43 | 172 | 10,4 | 8,062 | 9,422 | 116,9 | 0,643 | 6,8 | 0,132 | 1,4 |



ISO 23695 “ammonium nitrogen”

ILT results

- Within lab repeatability: 1.3–2.8 %
 - Same manufacturer
 - Same instrument
 - Same user
- Between lab reproducibility: 4.2–9.0%
 - Different manufacturers
 - Different instruments
 - Different users



Meets requirements of routine test



ISO 23696-1 / -2 “nitrate”





ISO 23696-1 / -2 “nitrate”

Why two parts?

- Chemistry
 - Part 1: Dimethylphenol
 - Part 2: Chromotropic Acid



Two established chemical indicators on the market



ISO 23696-1 / -2 “nitrate”

Composition information

Dimethylphenol

| Reagent name | Mass fraction range (% w/w) |
|--------------------|--------------------------------|
| 2,6-dimethylphenol | 0.007 to 1 |
| Sulfuric acid | 30 to 50 |
| Phosphoric acid | 25 to 50 |

Chromotropic acid reaction

| Reagent name | Mass fraction range (% w/w) |
|----------------------|--------------------------------|
| Sodium metabisulfite | 0.01 to 5 |
| Chromotropic acid | 0.01 to 1 |
| Sulfuric acid | 75 to 85 |



ISO 23696-1 / -2 “nitrate”

Procedure

NANOCOLOR® Nitrat 8
Nitrate / Nitrato

Methode / Method / Méthode / Método

| | | | | | |
|------|--|--------------------|------|------------------------------|--|
| 0651 | 0.30–8.00 mg/L NO ₃ -N | 345 / 350 / 365 nm | Min | 20–80 % | Max |
| 0652 | 1.3–35.0 mg/L NO ₃ ⁻ | | 0.30 | 1.84 | 6.46 |
| | | | 1.3 | 8.0 | 28.3 |
| | | | | 8.00 mg/L NO ₃ -N | 35.0 mg/L NO ₃ ⁻ |

Test 0-65
REF 985065

1 500 µL pH 1–13
2 500 µL R2
3 3 x
4 10 min
5
6 M
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A 037 171 / 985065 / xxxx
1/1
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ISO 23696-1 / -2 “nitrate”

ILT results for Part 1, dimethylphenol

| Sample | Matrix | I | n | o | X | \bar{x} | η | s_R | $C_{V,R}$ | s_r | $C_{V,r}$ |
|--------|---------------------|----|-----|------|-----------------|-----------------|--------|-----------------|-----------|-----------------|-----------|
| | | | | % | $\mu\text{g/L}$ | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % |
| A | Ground water | 46 | 184 | 4,2 | - | 1,32 | - | 0,132 | 10,0 | 0,040 | 3,0 |
| B | Swimming pool water | 39 | 156 | 17,0 | - | 8,42 | - | 0,166 | 2,0 | 0,077 | 0,9 |
| C | Waste water 1 | 45 | 180 | 0,0 | - | 50,29 | - | 1,302 | 2,6 | 0,485 | 1,0 |
| D | Waste water 2 | 41 | 164 | 10,9 | - | 14,79 | - | 0,511 | 3,5 | 0,151 | 1,0 |
| H | Surface water | 45 | 180 | 6,3 | - | 10,86 | - | 0,271 | 2,5 | 0,101 | 0,9 |
| I | QC Standard | 44 | 176 | 6,4 | 0,996 | 1,01 | 101,5 | 0,108 | 10,6 | 0,035 | 3,4 |



ISO 23696-1 / -2 “nitrate”

ILT results for Part 2, chromotropic acid

| Sample | Matrix | I | n | o | X | \bar{x} | η | s_R | $C_{V,R}$ | s_r | $C_{V,r}$ |
|--------|---------------------|----|----|------|-----------------|-----------------|--------|-----------------|-----------|-----------------|-----------|
| | | | | % | $\mu\text{g/L}$ | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % |
| A | Ground water | 10 | 40 | 0,0 | - | 1,55 | - | 0,279 | 18,0 | 0,099 | 6,4 |
| B | Swimming pool water | 10 | 40 | 0,0 | - | 8,77 | - | 0,640 | 7,3 | 0,198 | 2,3 |
| C | Waste water 1 | 7 | 28 | 12,5 | - | 52,85 | - | 2,647 | 5,0 | 0,887 | 1,7 |
| D | Waste water 2 | 10 | 40 | 0,0 | - | 15,39 | - | 0,399 | 2,6 | 0,212 | 1,4 |
| H | Surface water | 9 | 36 | 10,0 | - | 10,93 | - | 0,290 | 2,6 | 0,101 | 0,9 |
| I | QC Standard | 10 | 38 | 0,0 | 0,996 | 1,26 | 126,8 | 0,276 | 21,9 | 0,098 | 7,7 |



ISO 23696-1 / -2 “nitrate”

ILT results

- Dimethylphenol
 - Within lab repeatability: 0,9-3,4 %
 - Between lab reproducibility: 2,0-10,6%
 - Number of valid results: 39-46
- Chromotropic acid
 - Within lab repeatability: 0,9-6,4 %
 - Between lab reproducibility: 2,6-21,9%
 - Number of valid results: 9-10 (Waste water 1 = 7)



Meets requirements of routine test



ISO 23697-1 / -2 “total nitrogen”





ISO 23697-1 / -2 “total nitrogen”

Why two parts?

- Chemistry
 - Part 1: Dimethylphenol
 - Part 2: Chromotropic Acid



Two established chemical indicators on the market



ISO 23697-1 / -2 “total nitrogen”

Composition information

Digestion prior to Dimethylphenol detection of Nitrate

| Reagent name | Mass fraction range (% w/w) |
|----------------------|-----------------------------|
| Sodium hydroxide | 0.3 to 2 |
| Sodium carbonate | 0.8 to 1.3 |
| Potassium persulfate | 0.8 to 3 |
| Sodium sulfite | 0.15 to 0.3 |

Digestion prior to Chromotropic acid detection of Nitrate

| Reagent name | Mass fraction range (% w/w) |
|----------------------|-----------------------------|
| Sodium hydroxide | 0.3 to 2 |
| Potassium persulfate | 0.8 to 3 |
| Sodium metabisulfite | 0.01 to 5 |
| Chromotropic acid | 0.01 to 1 |
| Sulfuric acid | 50 to 70 |



ISO 23697-1 / -2 “total nitrogen”

Procedure – Part 1

NANOCOLOR® gesamt-Stickstoff TN_b 22
total Nitrogen / Azote total / Nitrógeno total

Test 0-83
REF 985083

Methode / Method / Méthode / Método 345 / 350 / 365 nm

| | Min | 20–80 % | Max |
|-----|-----|---------|-------------|
| 0.5 | 4.8 | 17.7 | 22.0 mg/L N |

① 5.0 mL
pH 5–9

②

③ 120 °C / 30 min
100 °C / 1 h

④ 20–25 °C

⑤

⑥

⑦ A

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1/2



ISO 23697-1 / -2 “total nitrogen”

Procedure – Part 2

NANOCOLOR® gesamt-Stickstoff TN_b 22
total Nitrogen / Azote total / Nitrógeno total

Test 0-83
REF 985083

| Methode / Method / Méthode / Método | 345 / 350 / 365 nm | Min | 20–80 % | Max |
|-------------------------------------|--------------------|-----|----------|-------------|
| 0831 0.5–22.0 mg/L N | | 0.5 | 4.8 17.7 | 22.0 mg/L N |

⑧ N22 500 µL A

⑨ 500 µL R2

⑩ 3 x

⑪ 10 min

⑫

⑬ M

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A037176 / 985083 / xxxx

2/2



ISO 23697-1 / -2 “total nitrogen”

ILT results for Part 1, dimethylphenol

| Sample | Matrix | I | n | o | X | \bar{x} | η | s_R | $C_{V,R}$ | s_r | $C_{V,r}$ |
|--------|---------------|----|-----|------|-----------------|-----------------|--------|-----------------|-----------|-----------------|-----------|
| | | | | % | $\mu\text{g/L}$ | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % |
| E | Waste water 3 | 41 | 164 | 10,9 | - | 16,30 | - | 0,864 | 5,3 | 0,281 | 1,7 |
| F | Waste water 4 | 45 | 180 | 4,3 | - | 61,63 | - | 3,526 | 5,7 | 1,530 | 2,5 |
| G | Waste water 5 | 45 | 179 | 4,8 | - | 70,42 | - | 9,996 | 14,2 | 2,341 | 3,3 |
| I | QC standard | 44 | 174 | 8,4 | 13,033 | 13,15 | 100,9 | 0,698 | 5,3 | 0,260 | 2,0 |



ISO 23697-1 / -2 “total nitrogen”

ILT results for Part 2, chromotropic acid

| Sample | Matrix | I | n | o | X | \bar{x} | η | s_R | $C_{V,R}$ | s_r | $C_{V,r}$ |
|--------|---------------|----|----|------|-----------------|-----------------|--------|-----------------|-----------|-----------------|-----------|
| | | | | % | $\mu\text{g/L}$ | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % | $\mu\text{g/L}$ | % |
| E | Waste water 3 | 10 | 40 | 0,0 | - | 1,55 | - | 0,279 | 18,0 | 0,099 | 6,4 |
| F | Waste water 4 | 10 | 40 | 0,0 | - | 8,77 | - | 0,640 | 7,3 | 0,198 | 2,3 |
| G | Waste water 5 | 7 | 28 | 12,5 | - | 52,85 | - | 2,647 | 5,0 | 0,887 | 1,7 |
| I | QC standard | 10 | 40 | 0,0 | - | 15,39 | - | 0,399 | 2,6 | 0,212 | 1,4 |



ISO 23697-1 / -2 “total nitrogen”

ILT results

- Dimethylphenol
 - Within lab repeatability: 1,7-3,3 %
 - Between lab reproducability: 5,3-14,2%
 - Number of valid results: 41-45
- Chromotropic acid
 - Within lab repeatability: 3,9-5,9 %
 - Between lab reproducability: 5,4-29,3%
 - Number of valid results: 9-11



Meets requirements of routine test



Ad-Block



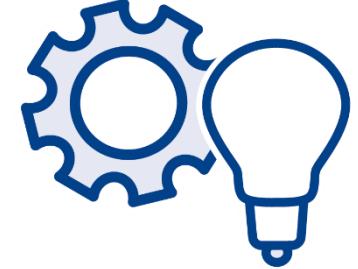
Ad Block



NANCOLOR Advance + Tube tests for all applications



MACHEREY-NAGEL – reliable water analysis



Summary





Summary

- Useful ISO norms are on their way
 - ISO 23695 Ammonium
 - ISO 23696 Nitrate
 - ISO 23697 Total Nitrogen
- Interlaboratory trial
 - Interlab reproducibility in general <15%
- If you want to test – use NANOCOLOR® by MN



MACHEREY-NAGEL – reliable water analysis

If you want to test – use NANOCOLOR® by MN

Dr. Christian Prokisch | cprokisch@mn-net.com | +49-2421-969-166

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