KVCV Studiedag
Trends in organische analyse

Erwin Van Poppel

Recente ontwikkelingen in extractietechnieken

DIONEX
Passion. Power. Productivity.

Recente ontwikkelingen in extractietechnieken
- Welke richting gaan we uit?

Presentatie overzicht

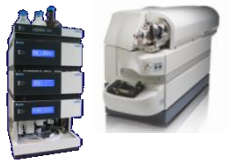
- 1-Het gebruik en de applicaties van Accelerated Solvent Extraction als monstervoorbereiding.
- 2-Geautomatiseerde Solid-Phase Extraction (SPE) met de AutoTrace® 280.
- 3-Online Solid Phase Extraction (SPE)

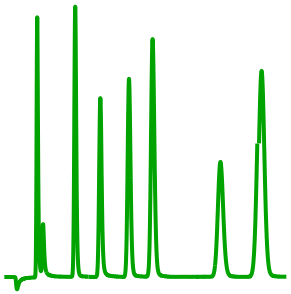
Sample Analysis Flow Path

Samples

Extraction

Separation
+
Detection





Data Analysis and Reporting

1-Het gebruik en de applicaties van Accelerated Solvent Extraction als monstervoorbereiding.

ASE® EPA Method 3545A

Polychlorinated Biphenyls

Semivolatiles

Perchlorate

Brominated Flame Retardants

Organophosphates

Chlorinated Herbicides

Chlorinated Dioxins and Furans

What Is ASE®?

- ASE automates sample preparation for **solid** samples using solvent extraction
- ASE typically operates above the boiling point of most extraction solvents
- Pressure is used to keep solvents liquid during extraction
- ASE is equivalent or superior to Soxhlet extraction

ASE is approved as US EPA Method 3545a

ASE® Schematic

Cell loading

Fill, heat, and equilibrate — Total (min) 5 to 9

dynamic extraction

Static extraction — 0 to 99

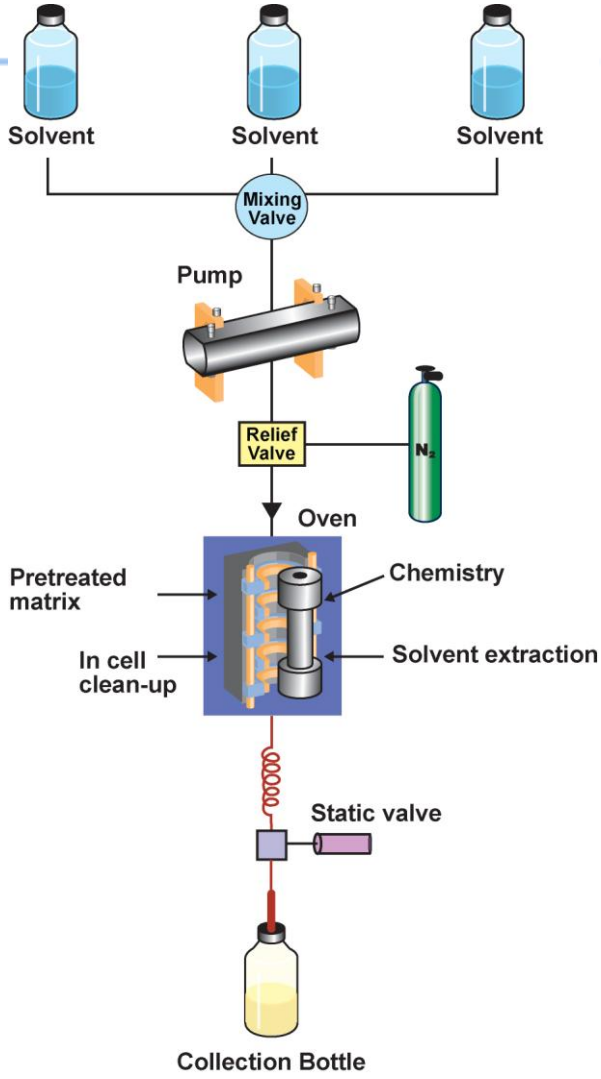
cycle

Rinse with fresh solvent — 0.5

static extraction

Purge with nitrogen — 1–2

Extract ready — Total (min) 12–20



ASE® Introduced in 1995

- Speed: extract in 20 minutes
- Efficiency: recoveries equal or better than Soxhlet extraction
- Economy: low solvent usage
- Experience: 15 years, 4000+ systems, 600+ publications
- Many Official Methods
- ASE 150
- ASE 350

ASE® 350: The New Standard

- Combination of ASE 200 and 300 capabilities
  - 24 smaller cells (1, 5, 10, 22, or 34 mL)
  - 19 larger cells (66 or 100 mL)
  - 19 smaller cells and 5 larger cells
  - 60- or 250-mL collection bottles (up to 28 small and 5 large)
- Faster pump (70 mL/min versus 35 mL/min)
- Integrated solvent controller

Dionex ASE® 150 and ASE 350 Instruments

Summary of EPA Equivalency Study for ASE®

Compound Class	Comparison Technique	Relative Recovery
Organochlorine Pesticides (OCP)	Automated Soxhlet	97.3%
Organophosphorus Pesticides (OPP)	Automated Soxhlet	99.2%
Semivolatiles (BNA)	Soxhlet	98.6%
Chlorinated Herbicides	Shake Method	112.9%
Polychlorinated Biphenyls (PCB)	Various Reference Materials	98.2%
Polycyclic Aromatic Hydrocarbons (PAH)	Various Reference Materials	104.8%

- **ASE is similar to existing extraction techniques**

ASE® Environmental Applications

- US EPA method 3545A
  - Pesticides and herbicides
  - PAHs and semi-volatiles
  - PCBs
  - Dioxins and furans
  - TPH (Total Petroleum Hydrocarbons)
  - Explosives
- Air samples
  - PUF (polyurethane foam) cartridges
  - Quartz filters
  - XAD and charcoal
- PBDEs in flora, fauna, sediments and soils
- Method 6860
  - Perchlorate in soils, plants and animal tissues

ASE® Pharmaceutical Applications

- Natural products from plants
  - Raw materials testing
  - High throughput screening
- Active ingredients
  - Patches, tablets and animal feeds
- Herbal supplements
  - St. John's Wort
  - Goldenseal Root
  - Kava
  - Ephedra
- Parent compound and metabolites in tissues

ASE® Applications for Consumer Products

- Active ingredients in OTC medications
  - Ointments, creams
- Textiles
- Extractables in detergents
- Pulp and paper
- Extractable in roofing materials

ASE® Food Applications

- Contaminant analysis
  - Pesticides
  - Herbicides
  - PCBs, dioxins, etc.
- Component analysis
  - Lipid content
  - Flavors, aromas, etc.

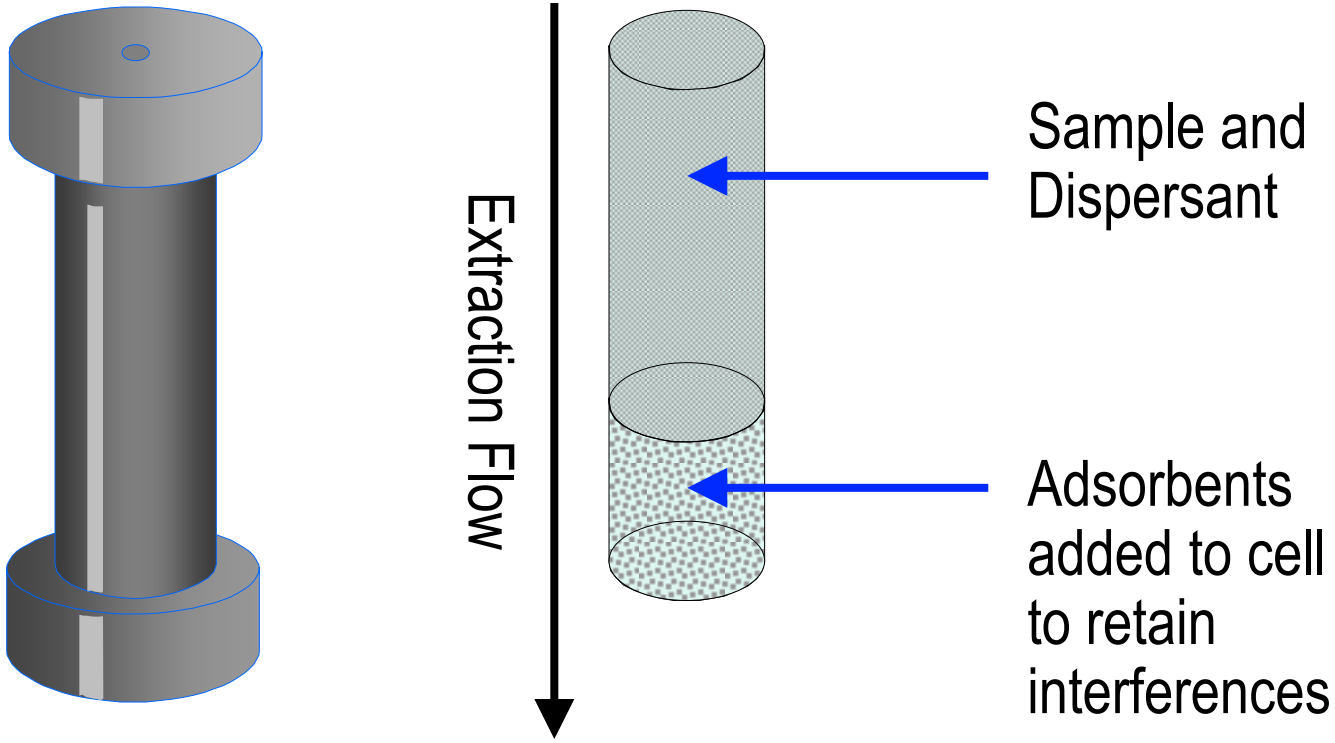
Contaminants in Food Matrices Evaluated with ASE®

- Pesticides in fruits and vegetables
- Antibiotics in animal tissue
- Dioxins and PCBs in animal tissue
- Pesticides and herbicides in wheat
- Mycotoxins in grain
- PAH in smoked meats
- Nitrate and nitrite in bacon
- Acrylamide in food (Dionex AN 358 and 406)

Integrated Clean-Up
Salmon Extracts

Extracts With and Without In-Cell Clean-Up of Fish Tissue Using Alumina, Silica Gel, and Acidic Silica Gel (40% H_2SO_4)

Dionex Tools and Techniques for Sample Preparation



Schematic of In-Cell Cleanup with ASE®

Dionex Tools and Techniques for Sample Preparation

Absorbents	*Interferences*	*Analyte Group*
Carbon	Organics	Non-polar compounds, dioxins
Copper	Elemental sulfur	Multi-residue pesticides
Ion-exchange resins	Organics, metals and ionic interferences	Anions, cations, metal speciation (arsenic)
C18 resins	Organics, lipids, chlorophyll	Non-polar compounds
Acid impregnated silica gel	Lipids and oils	PCB and bromated flame retardants
Alumina	Lipids, chlorophyll, petroleum, waste	Amines, perchlorates, and PCB's
Florisil™	Oils, lipids, and waxes	Pesticides and aromatics

Schematic of In-Cell Cleanup with ASE®

ASE® of Cr(VI) from Soil

- Add sample to an ASE cell
- Add 0.5 mL of 1.0 M phosphate buffer
- Solvent: ammonium sulfate buffer, pH 9-9.5
- Temperature/pressure: 200 °C/10.3 MPa
- Static cycles/time: 4 x 7-min
- Speed: 44 min
- Economy: 50 mL solvent

Analysis by Dionex IC (DX-600) EPA method 7199 conditions for IC

Comparative Results for Cr(VI)

Sample	ASE® and EPA 7199	EPA 3060A and EPA 7199	Colorimetric method
Site A	5mg/Kg	7 mg/Kg	8mg/Kg
Site B	70mg/Kg	67 mg/Kg	90 mg/Kg
Site C	235 mg/Kg	177 mg/Kg	200mg/Kg

Data courtesy of Franco Abballe, Dionex Italy

Aarde, Wind en Water

2-Geautomatiseerde Solid-Phase Extraction (SPE) met de AutoTrace® 280.

- Accelerated Solvent Extraction is great for solids: soils, foods polymers animal and plant tissues
- For years customers have asked ”when will you make something for water samples”?
- Large volume Automated Solid phase extraction for organic analytes from water

AutoTrace® SPE Work Station

AutoTrace provides reliable automated SPE for analytical chemists determining organic pollutants in large-volume water samples. Unlike traditional methods such as liquid-liquid extraction using separatory funnels, AutoTrace saves time, solvent and labor ensuring high reproducibility and productivity for analytical laboratories. The unit can process up to 6 samples in 2-3 hours. The AutoTrace uses powerful pumps and a proven positive pressure technology to efficiently process and the most difficult samples.

AutoTrace® Background

- AutoTrace was introduced in 1992 by Zymark
  - Large volume solid phase extraction (SPE)
  - Zymark also manufactured TurboVap and Benchmate
  - Caliper Life Science and Zymark merger in 2003
- November 2008 Dionex purchase AutoTrace product from Caliper Life Science
  - Product, trademarks, existing customer base
- Dionex begins taking orders January 2, 2009
- Manufacturing begins in Sunnyvale, February 1, 2009

AutoTrace® 280 SPE Instrument

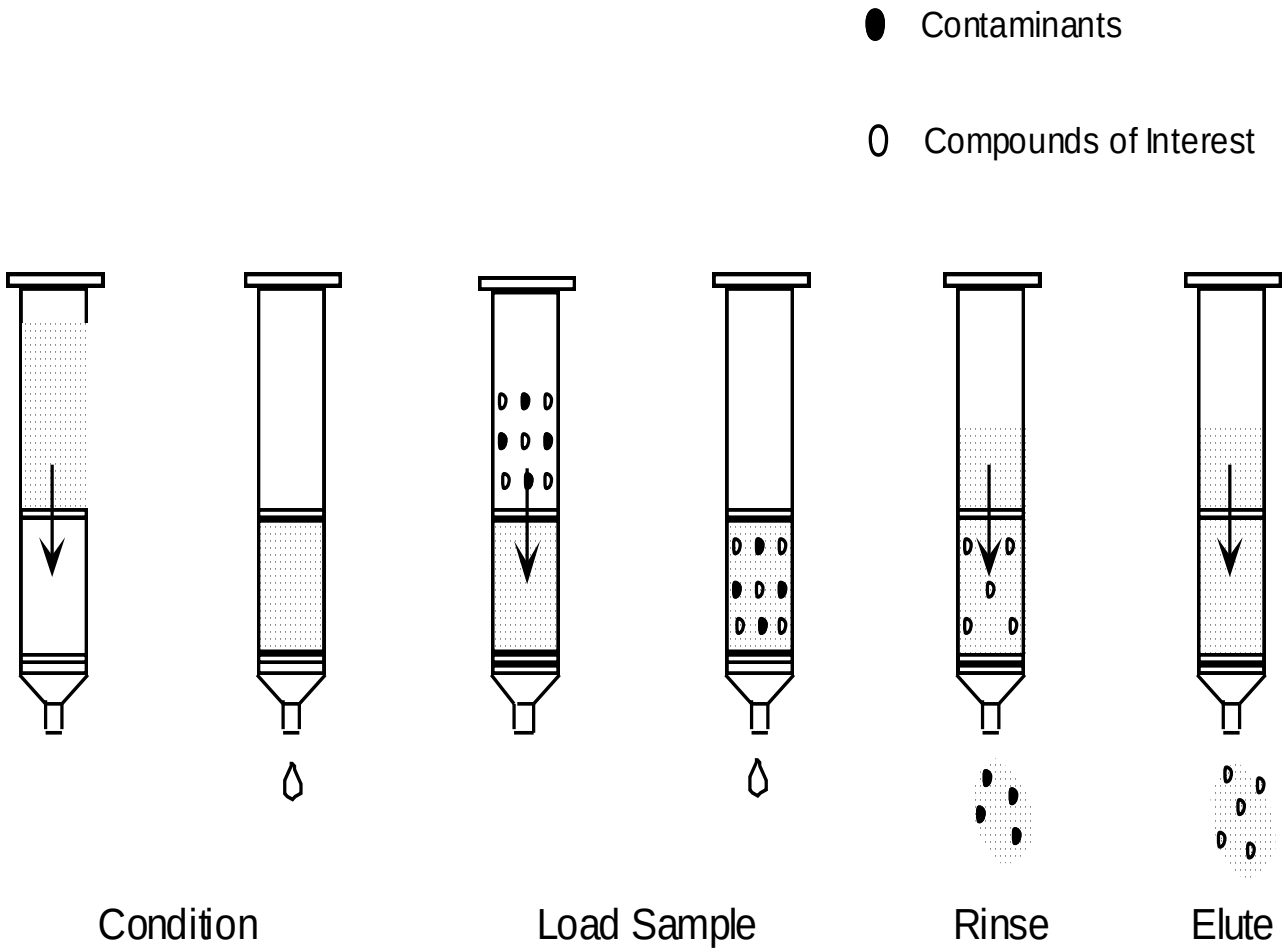
What is AutoTrace®?

- Automated solvent extraction of large-volume water samples
  - 20 mL to 20 L
  - Drinking water and groundwater
  - Wastewater may require pre-filtration
- Sample prep for organic analytes
  - Priority organic pollutants, personal care products, and endocrine disruptors
- Automated Solid-Phase Extraction (SPE)
  - Condition, load, rinse, and elute
  - Use normal or reversed-phase cartridges and disks
    - 1, 3, and 6 mL cartridges and 47 mm disks

SPE Terminology

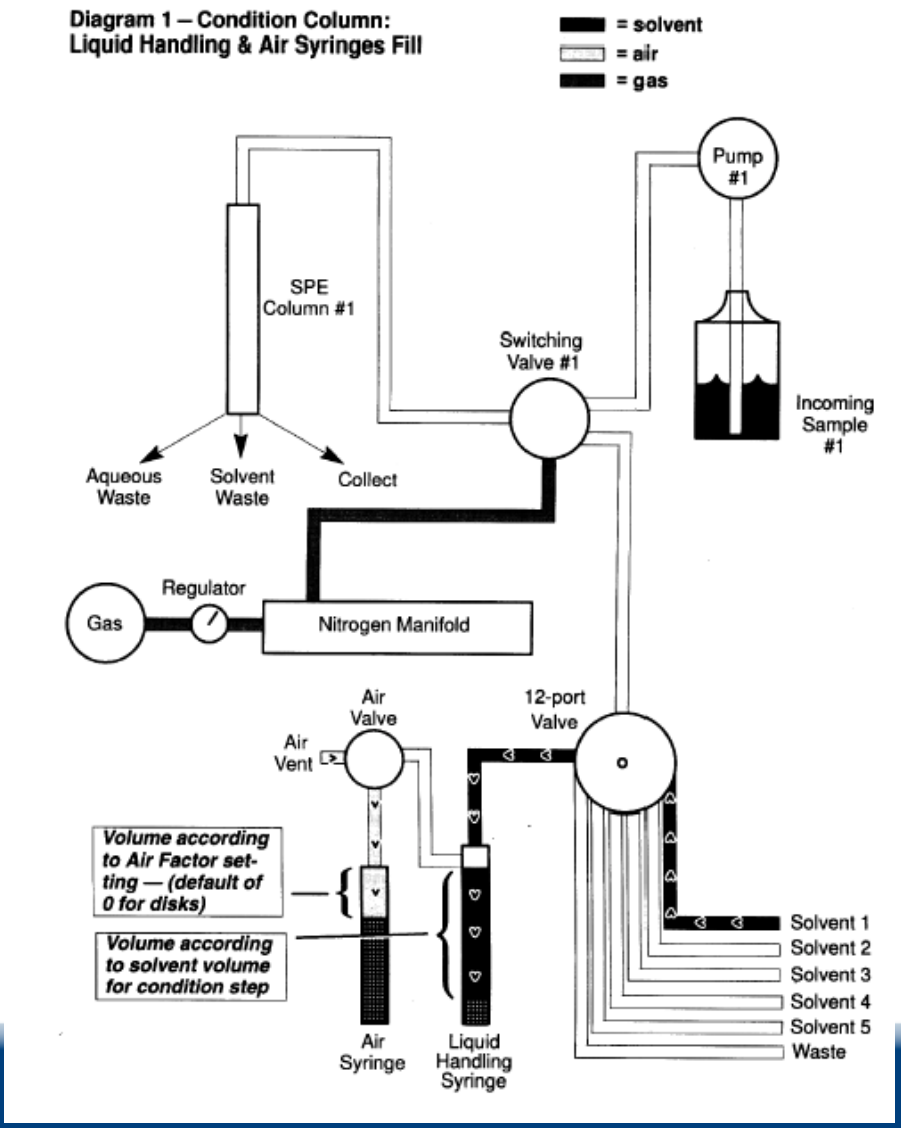
- **Condition:** Reagents are used to activate the functional groups on the column so the analyte(s) can bind to the column.
- **Load:** A sample is loaded on to the SPE column. If the analytes of interest bind to the column the sample would be "Loaded to Waste." If the analytes of interest pass through the column the sample would be "Loaded to Collect."
- **Rinse:** Rinse reagents are used to remove unwanted materials retained on the column.
- **Elute:** An elution reagent is used to remove the analytes of interest from the SPE column.

The Mechanics of SPE



AutoTrace® 280 SPE Instrument

AutoTrace® Operation



AutoTrace® 280 Features and Specifications

- Sample size 20 mL–20 L
  - Manual rinse of sample bottle
- 1, 3, and 6 mL cartridges
- 47 mm disks
- Sample loading in parallel
  - Up to 30 mL/min cart
  - Up to 60 mL/min disk
- Condition, rinse, and elute six samples sequentially
- Sample and solvents travel in different tubing to cartridge
  - Competitors use same path

AutoTrace® 280 Features (continued)

- Many different collection or elution container configurations
  - 16 × 100 mm test tubes
  - 17 × 60 mm vials
  - 11 mm GC vial
  - 4 mL WISP vial
  - 15 mL centrifuge tube

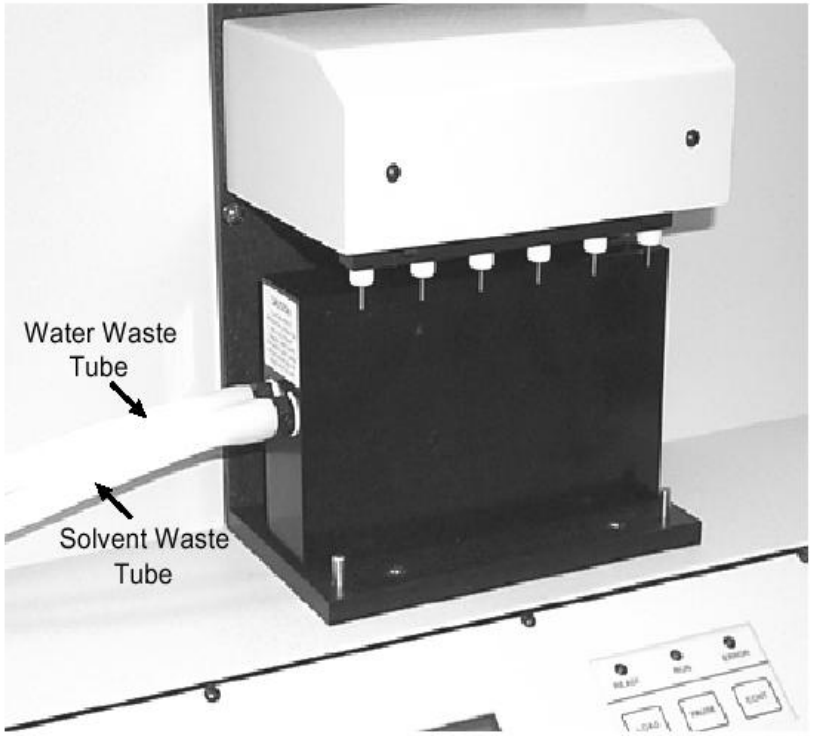


Figure 7. Waste Tubing Connections

AutoTrace® Benefits

- SPE uses less solvent when compared to traditional extractions
- Positive pressure for SPE functions
  - Other systems use a vacuum
- Automates all four SPE steps
  - Condition, load, rinse and elute
- Segregates aqueous and solvent waste using separate waste lines
- Processes ground, surface and wastewater samples
- Easy to use SPE specific software
- Documented EPA methods for water testing
- Integrated aspiration for solvent vapors: No need for a hood

Applications for the AutoTrace® 280 Instrument

1. Phenols, PAH'S, PCB'S, Dioxins
2. Organophosphorus and organochlorine pesticides
3. Nitrosamines and acid herbicides
4. Urons (diurons–weed killers)
5. Diquat in drinking water
6. Steroids, estrogen and endocrine disruptors
7. Pharmaceuticals and explosives

Application notes are available at www.dionex.com

Pesticide Recovery Study

Table 1

Pesticide Recovery Study
AutoTrace SPE Workstation vs. Vacuum Manifold SPE

	AutoTrace SPE		Vacuum Manifold SPE	
Compound	Recovery%	RSD%	Recovery%	RSD%
Atrazine	88	1.8	54	12.2
Propazine	91	1.5	80	7.3
Alachlor	99	3.4	96	4.1
Metalachlor	99	4.3	96	2.9
N=6				

EPA 505—OCP Method for SPE Disk Method

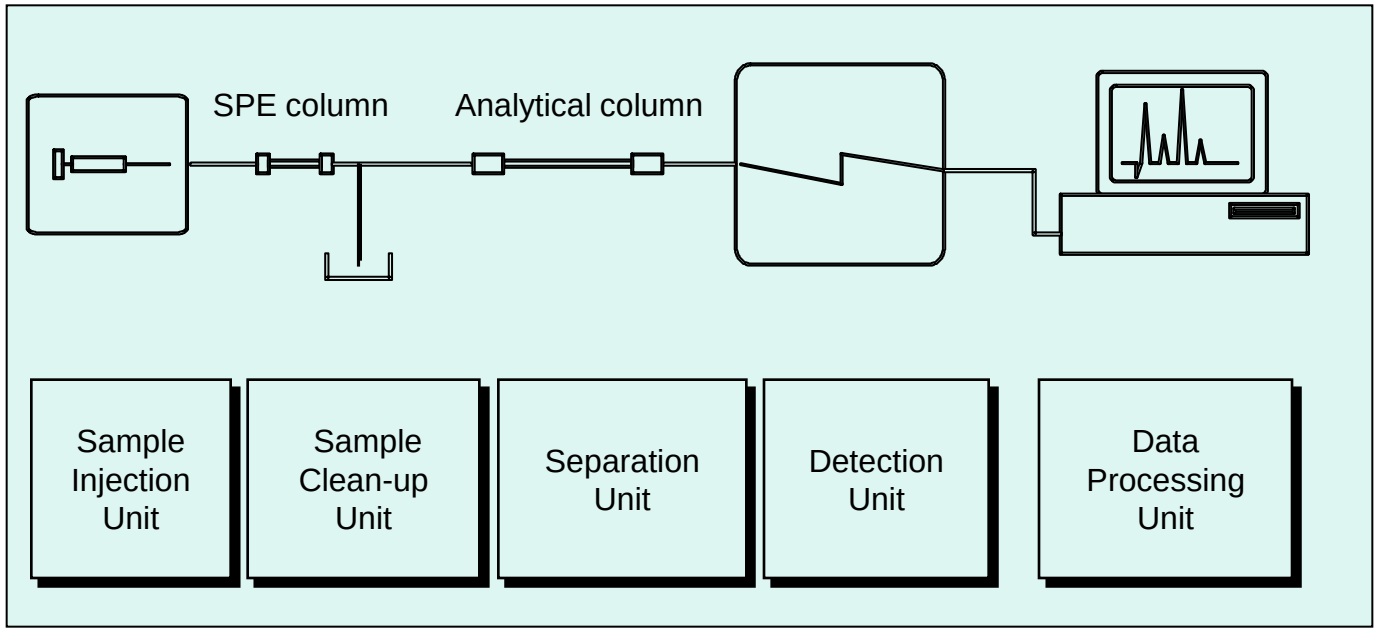
- Condition and Load Method Steps
  - Condition disk with 2.0 mL of CH_3OH
  - Rinse disk with 5.0 mL of ethyl acetate
  - Rinse disk with 5.0 mL of CH_2Cl_2
  - Condition disk with 10.0 mL of CH_3OH
  - Condition disk with 10.0 mL of pH 2 water
  - Load 120.0 mL of sample onto disk
  - Dry disk with nitrogen gas
- Elution Method Steps
  - Rinse sample container with 10.0 mL ethyl acetate to collect
  - Rinse sample container with 20.0 mL CH_2Cl_2 to collect
  - Soak and collect 2.0 mL fraction using ethyl acetate
  - Soak and collect 3.0 mL fraction using CH_2Cl_2

Results of EPA 505—Disk Method

Peak Name	Amount (µg/L)	% Recovery	%RSD
Hexachlorocyclopentadiene	45.1	90.2	3.7
Hexachlorobenzene	48.6	97.3	3.3
Pentachloronitrobenzene	53.6	107.2	3.3
Aldrin	48.9	97.8	4.1
Heptachlor epoxide	54.8	109.5	3.3
Gamma-Chlordane	50.4	100.8	3.6
Alpha-Chlordane	50.4	100.8	3.6
trans-Nonachlor	51.9	103.9	3.7
Dieldrin	52.6	105.3	3.3
Endrin	55.0	110.0	4.0
cis-Nonachlor	51.3	102.6	3.8
Methoxychlor	54.8	109.7	3.8

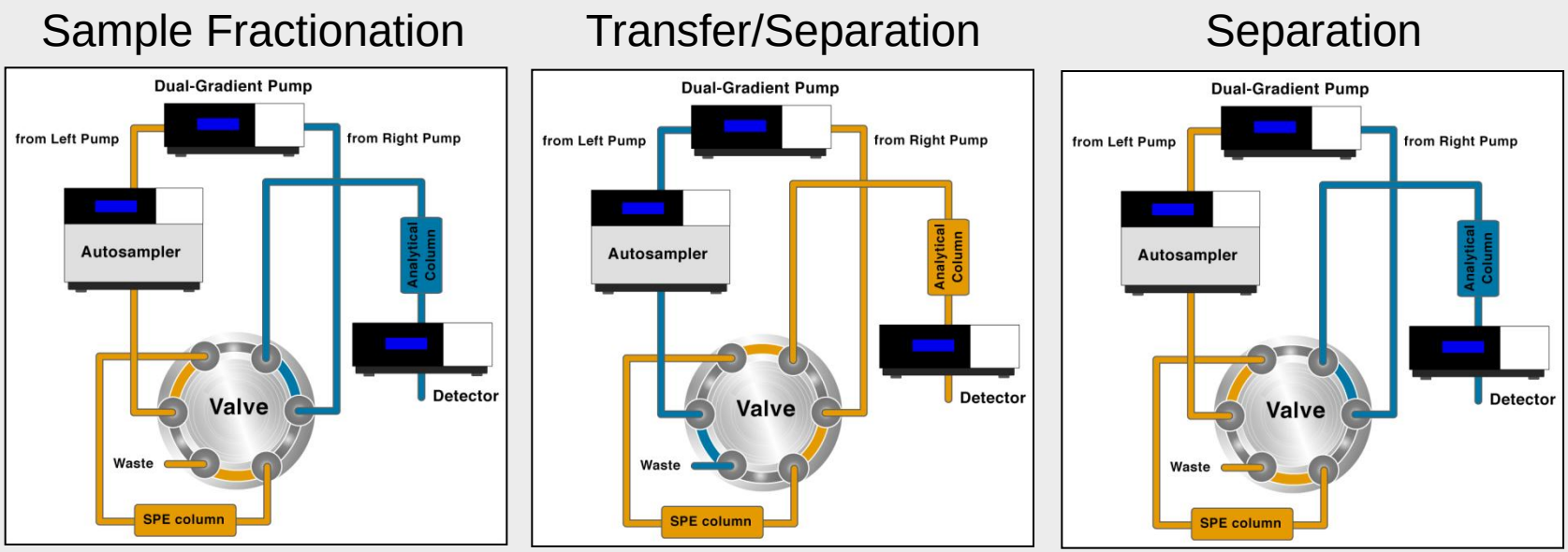
Results from 12 runs

3- Online Solid Phase Extraction (SPE)
Automated Sample Preparation - On-line SPE-LC



Total Analysis System (TAS) for on-line SPE

Instrumental Setup for On-line SPE-LC



- First dimension SPE column separates analytes from matrix
- After transfer to an analytical column, analytes are separated and detected
- SPE column can be washed and equilibrated in parallel to the analytical run

Convenient, fully automated sample preparation for complex samples

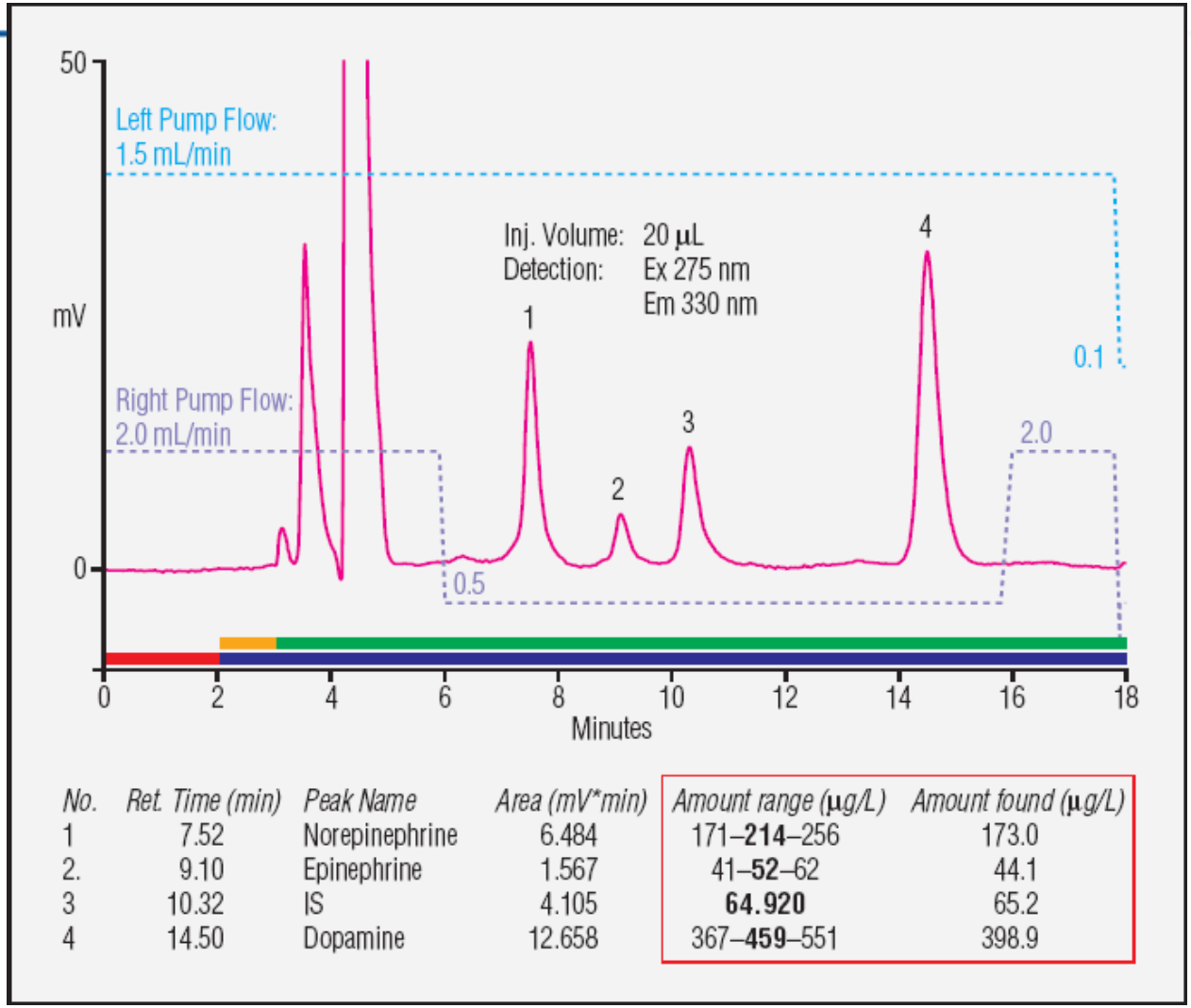
Automated Sample Preparation – Key Benefits

- Convenient direct injection of samples
- Sample preparation is highly reproducible
- Enables unattended operation, over night and over the weekend
- Reduces need for manual labor, frees operator time
- Minimizes contact with biologically hazardous samples
- Fast and cost-effective

Application areas:

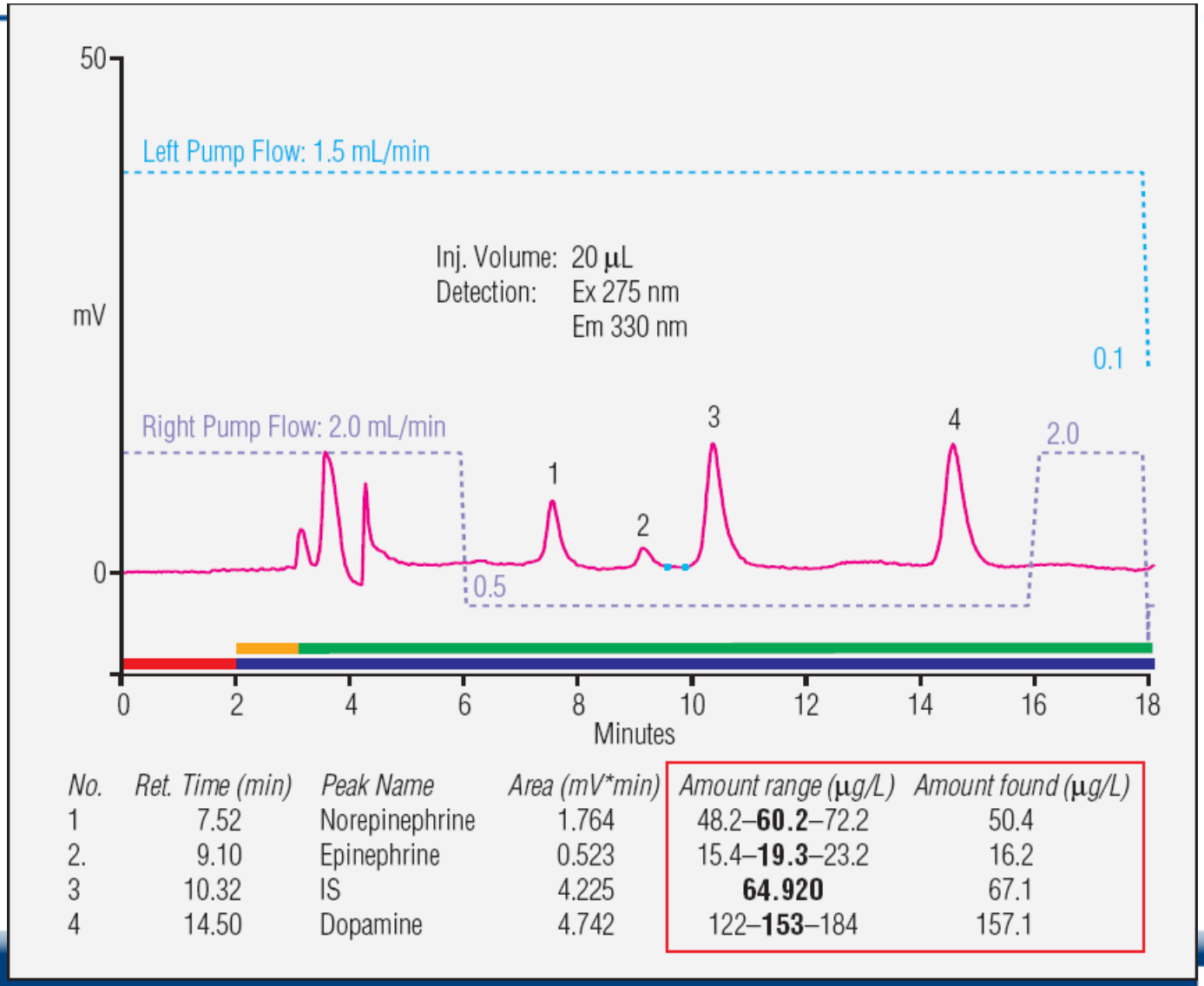
- Pharmaceutical and clinical labs: biological fluids
- Food and beverage labs
- Environmental labs

On-line SPE-LC-FD of Pathological Control Urine



No.	*Ret. Time (min)*	*Peak Name*	*Area (mV*min)*	*Amount range (µg/L)*	*Amount found (µg/L)*
1	7.52	Norepinephrine	6.484	171–**214**–256	173.0
2.	9.10	Epinephrine	1.567	41–**52**–62	44.1
3	10.32	IS	4.105	**64.920**	65.2
4	14.50	Dopamine	12.658	367–**459**–551	398.9

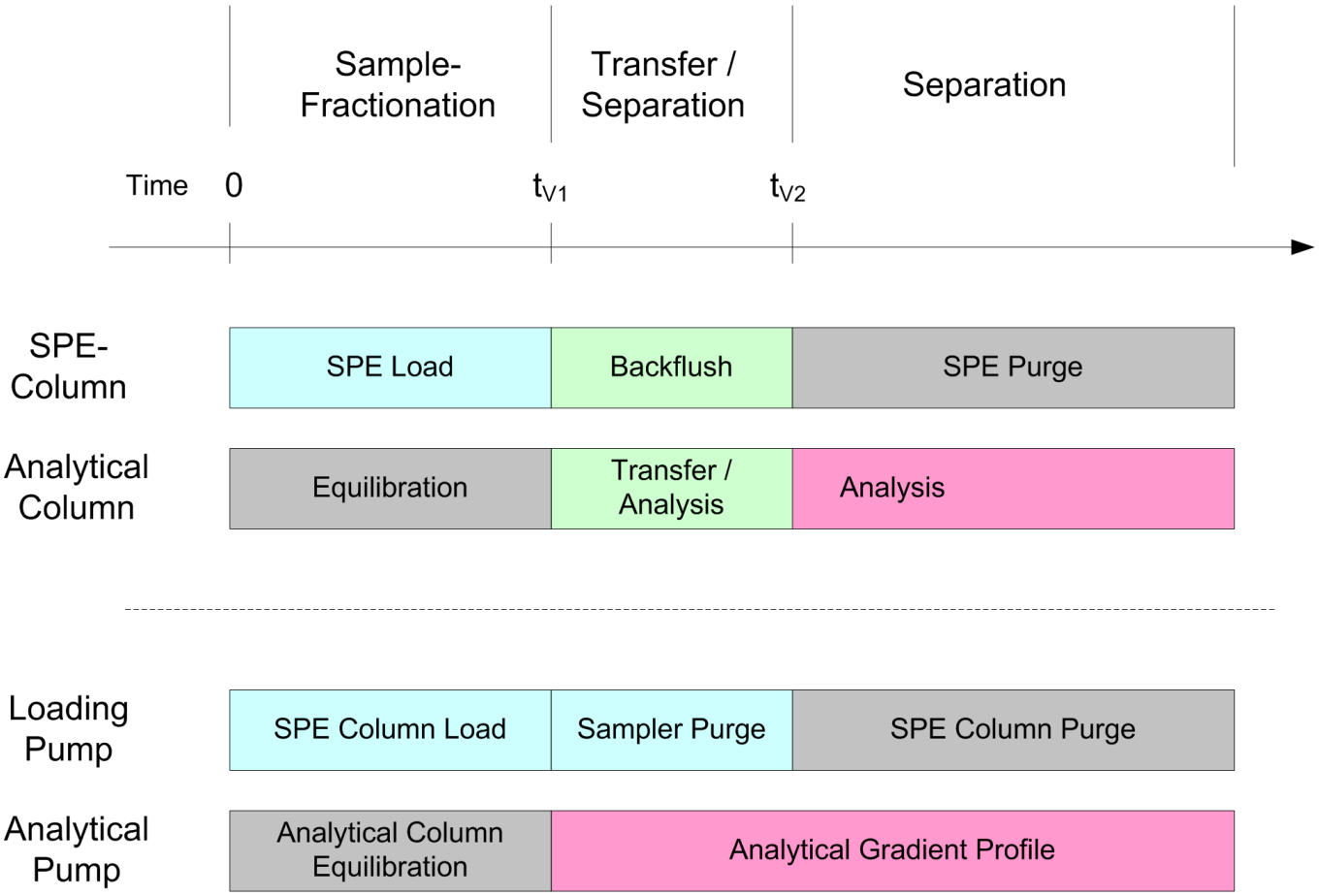
On-line SPE-LC-FD of Normal Control Urine



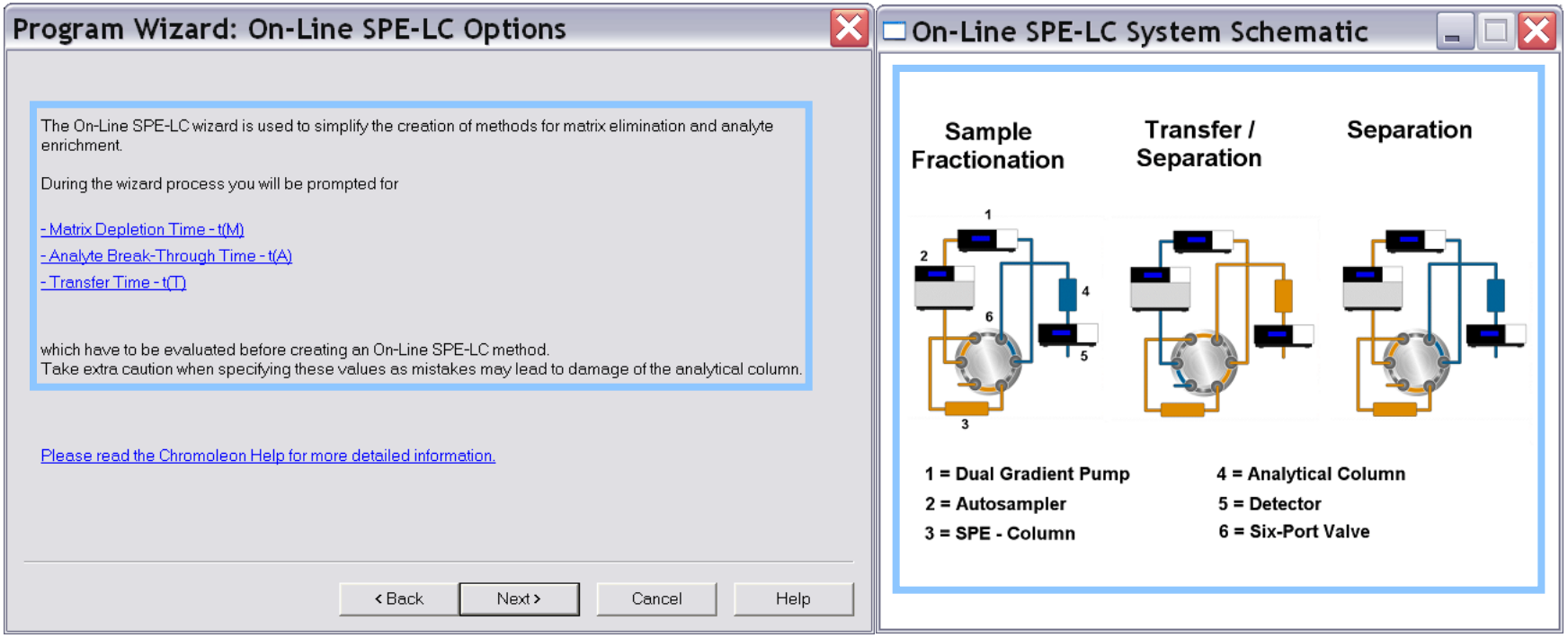
No.	Ret. Time (min)	Peak Name	Area (mV*min)	Amount range (µg/L)	Amount found (µg/L)
1	7.52	Norepinephrine	1.764	48.2–**60.2**–72.2	50.4
2.	9.10	Epinephrine	0.523	15.4–**19.3**–23.2	16.2
3	10.32	IS	4.225	**64.920**	67.1
4	14.50	Dopamine	4.742	122–**153**–184	157.1

DIONEX
Passion. Power. Productivity.

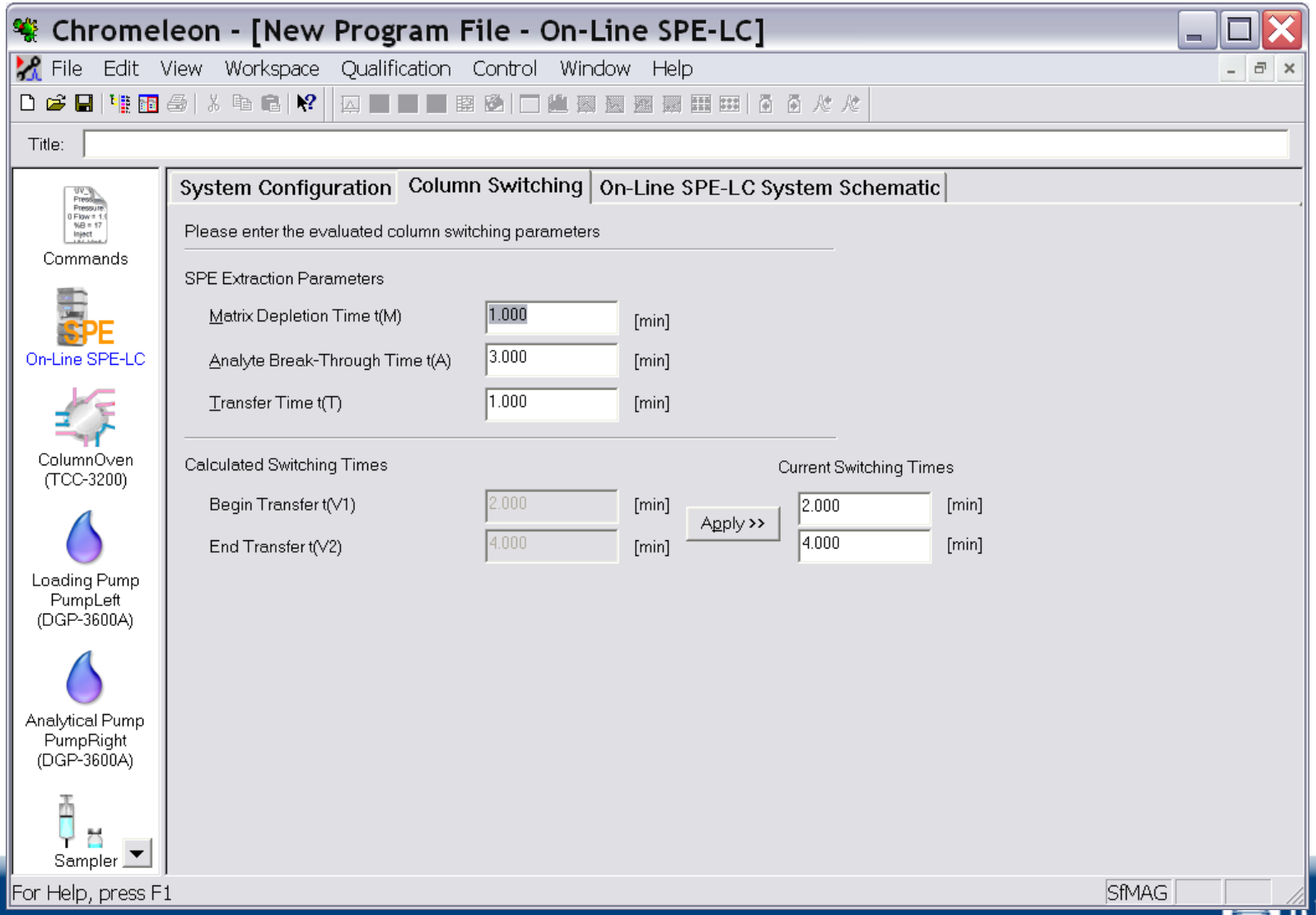
SPE Process Steps



On-line SPE-LC Software Wizard



Easy Editing of Valve Switching Times



DIONEX
Passion. Power. Productivity.

Thank you!

DIONEX

Passion. Power. Productivity.