Automated Concentration and Quantitative Transfer to Reduce Sample Handling

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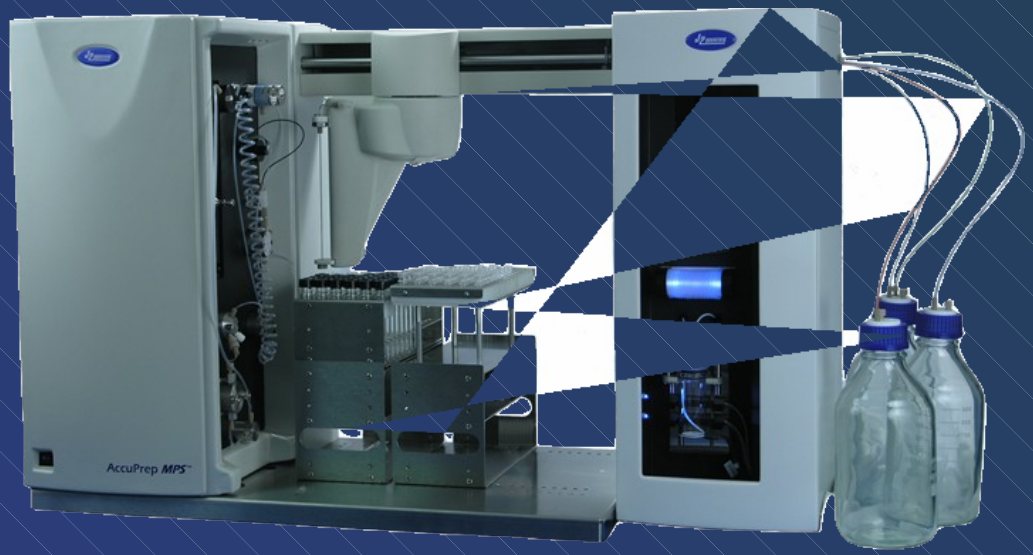
Abstract

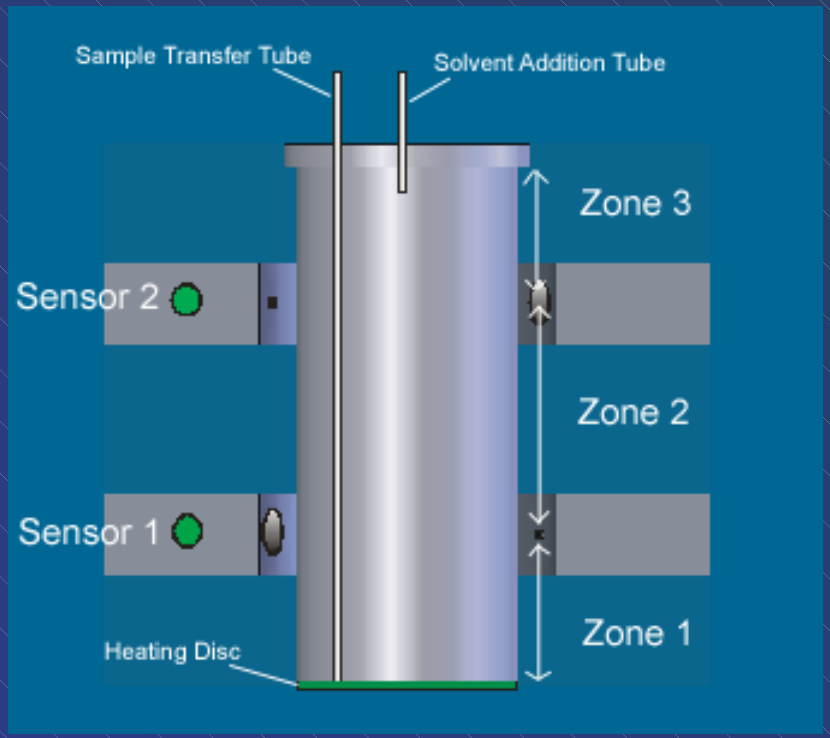
- Sample preparation often includes one or more concentration steps. Traditional systems vary in level of automation and final volume precision.
- Fully automated system offers flexible platform to meet needs of laboratories.
  - Capital investment = reduced per-sample cost
  - Less handling = more consistent recoveries

Instrumentation



AccuPrep MPS GPC Cleanup with
AccuVap Online Evaporation System
from J2 Scientific

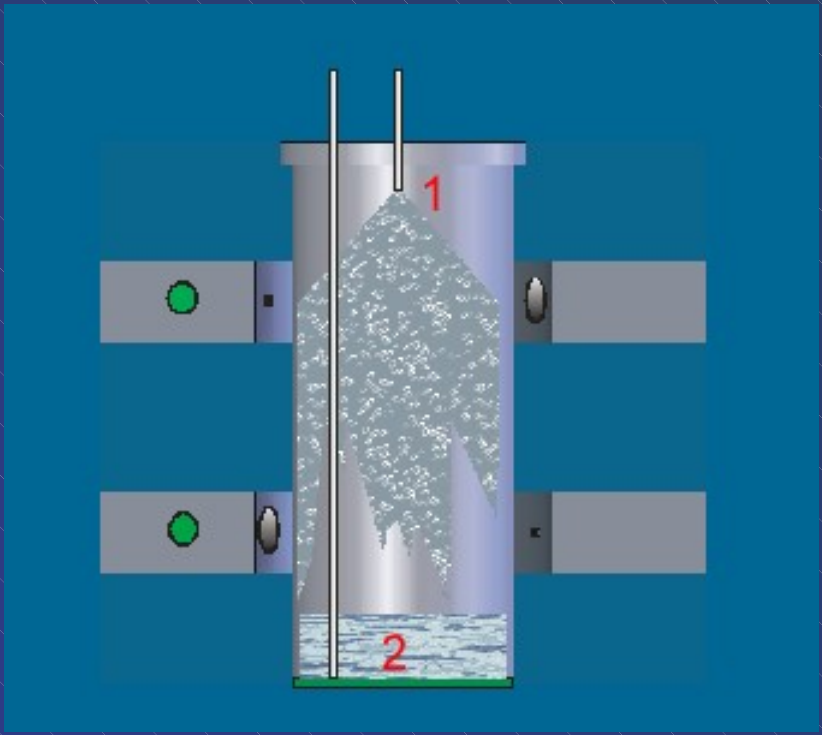
Evaporation Chamber Control



- Temperature & vacuum independently set for each zone.
- Actual vacuum & temperature values monitored by sensors for precise feedback and control.
- Heat supplied by proprietary disc (bottom of chamber).

Evaporation Time

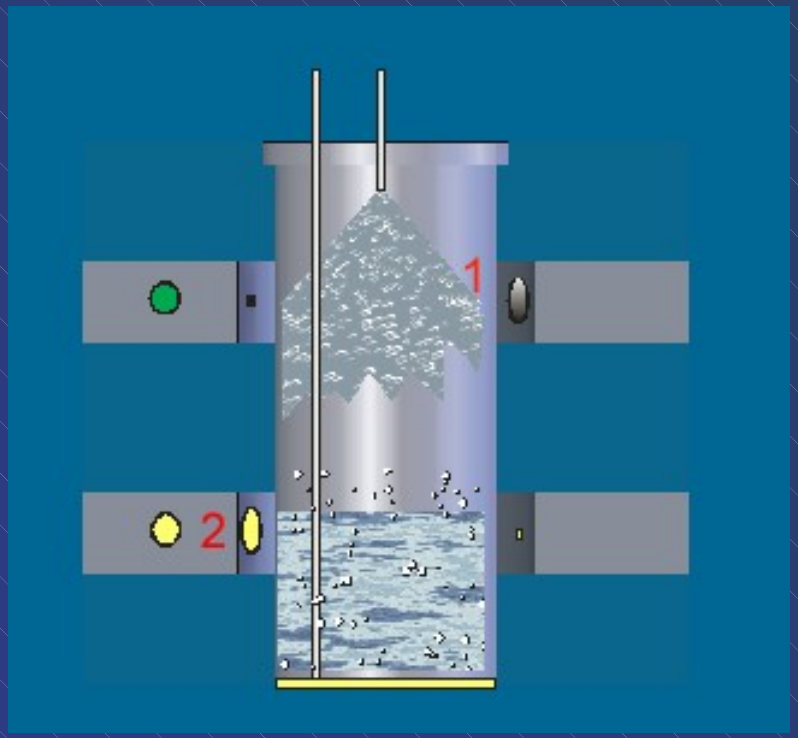
Detail 1



1. Sample aliqout added to chamber each 20 seconds as it elutes from column
2. As sample fluid accumulates, vacuum & heat are such that minimal evaporation occurs below Sensor 1 (prevents first additions from boiling dry)

Evaporation Time

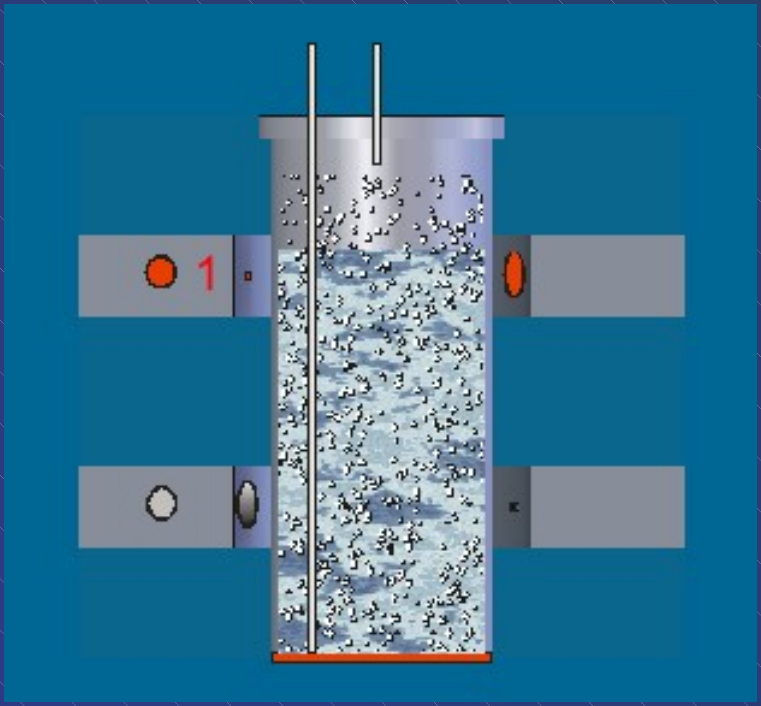
Detail 2



1. Sample continues to fill chamber, sprayed down sides to facilitate continuous rinsing of walls.
2. When level reaches Sensor 1, second programmed set of heat and vacuum values is activated. Level of sample stays between Sensor 1 and Sensor 2 during evaporation.

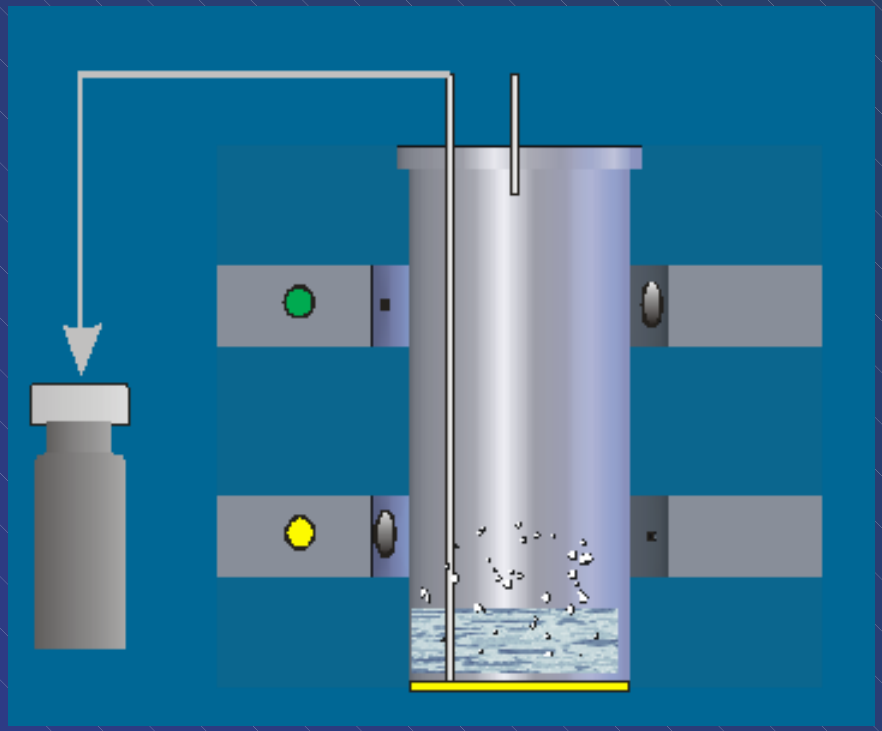
Evaporation Time

Detail 3



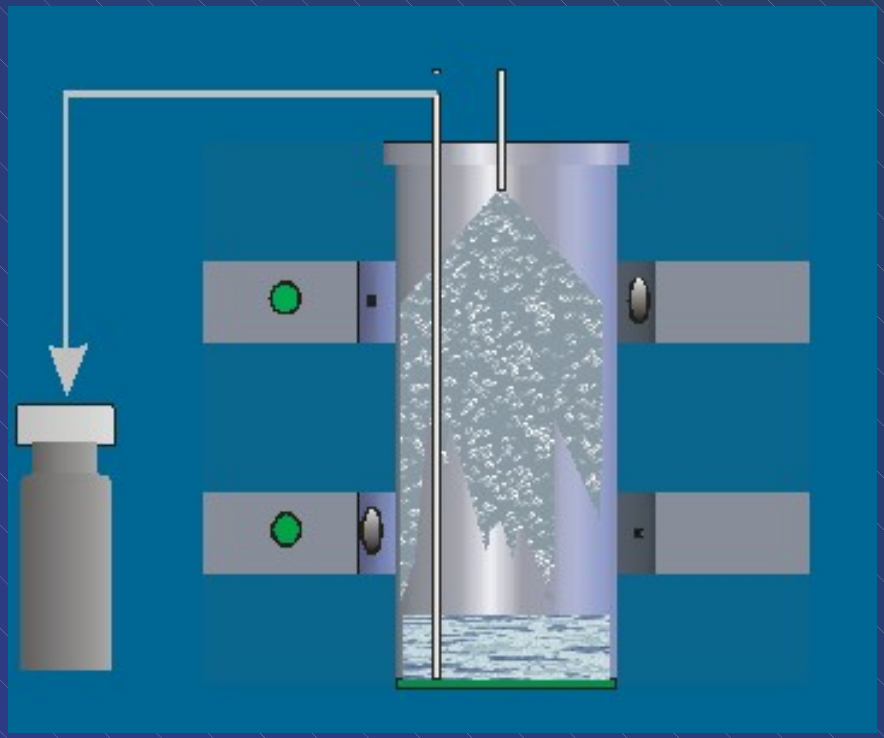
If solvent level in chamber reaches Sensor 2, third set of heat and vacuum parameters is activated, bringing sample level down and avoiding overfilled chamber

Endpoint Time:
Dryness 1



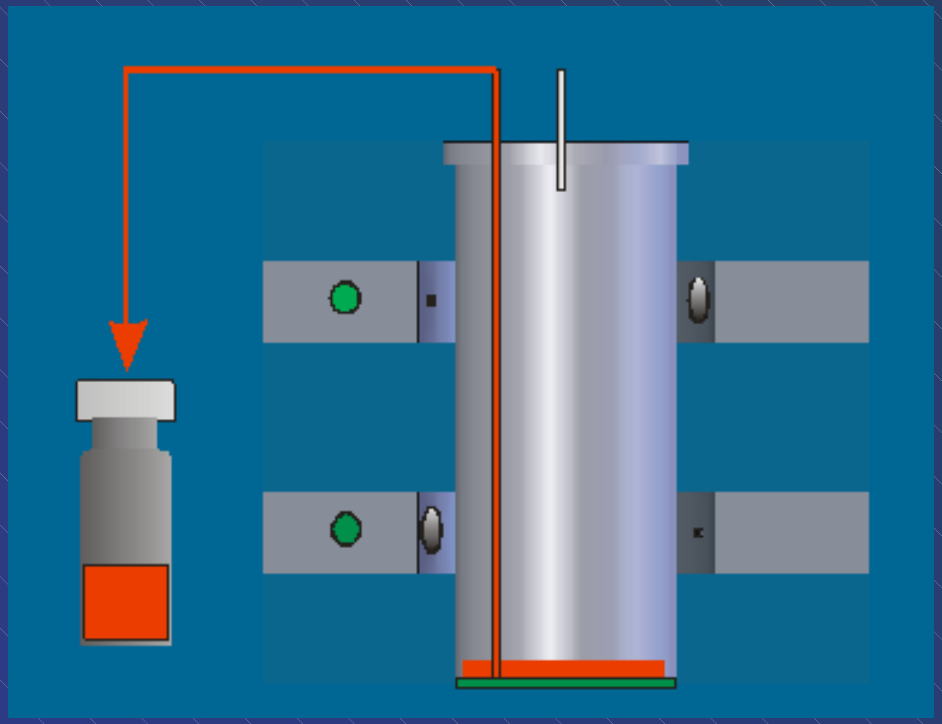
- After entire sample introduced to chamber (collect time complete) endpoint phase begins.
- End Pt choice 1: Dryness. Sample gently taken to dryness using programmed heat and vacuum settings to eliminate analyte loss.
- Dryness detected.
- Chamber is cooled prior to diluent addition.

Endpoint Time:
Dryness 2



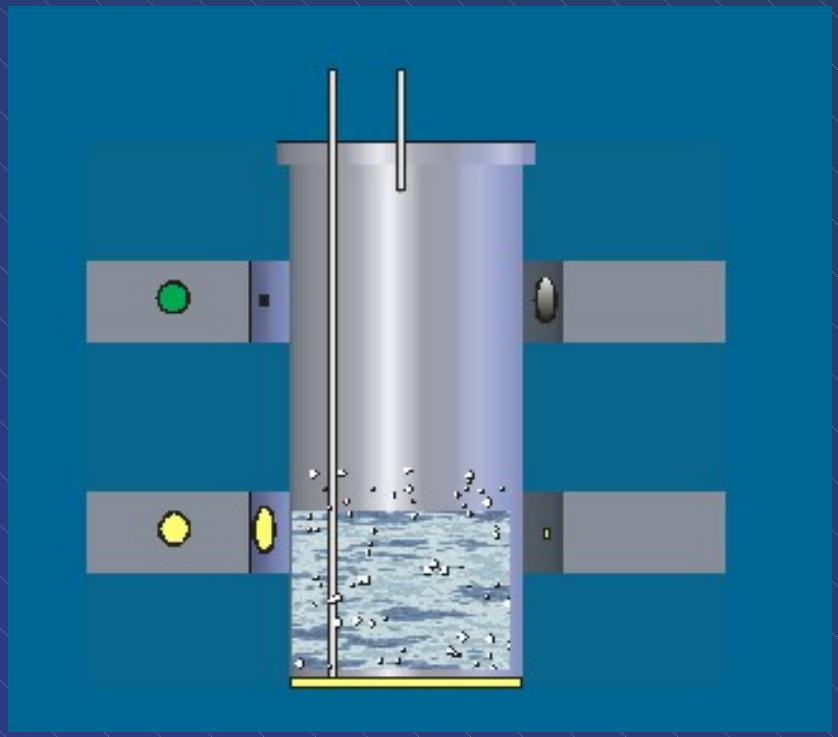
- After dryness is reached, programmed volume of diluent solvent is added to chamber and mixed using syringe pump.

Endpoint Time:
Dryness 3



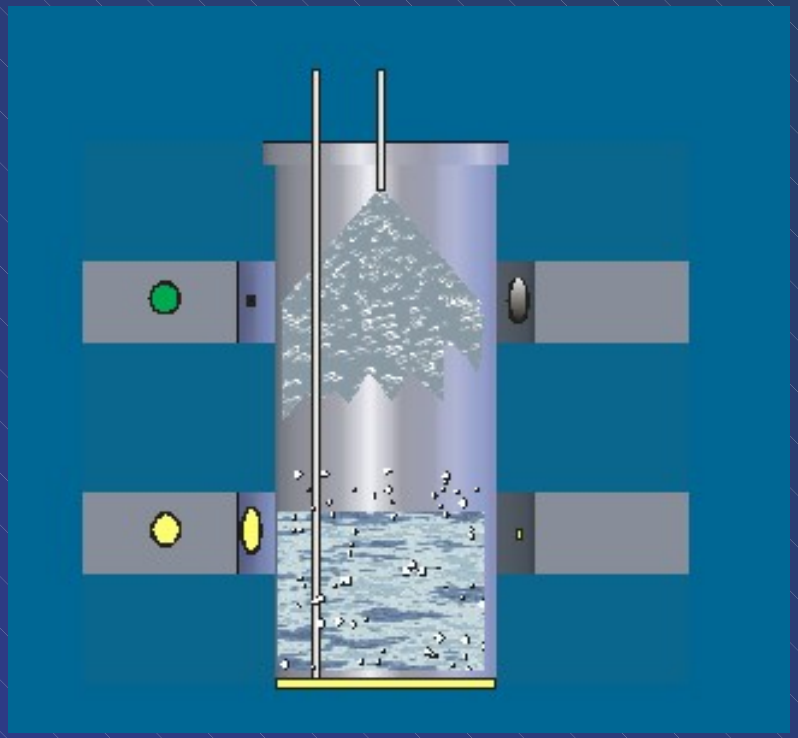
- After mixing, sample aliquot is accurately and precisely transferred to the collection vial using syringe pump

Endpoint Time:
Level Sensor 1



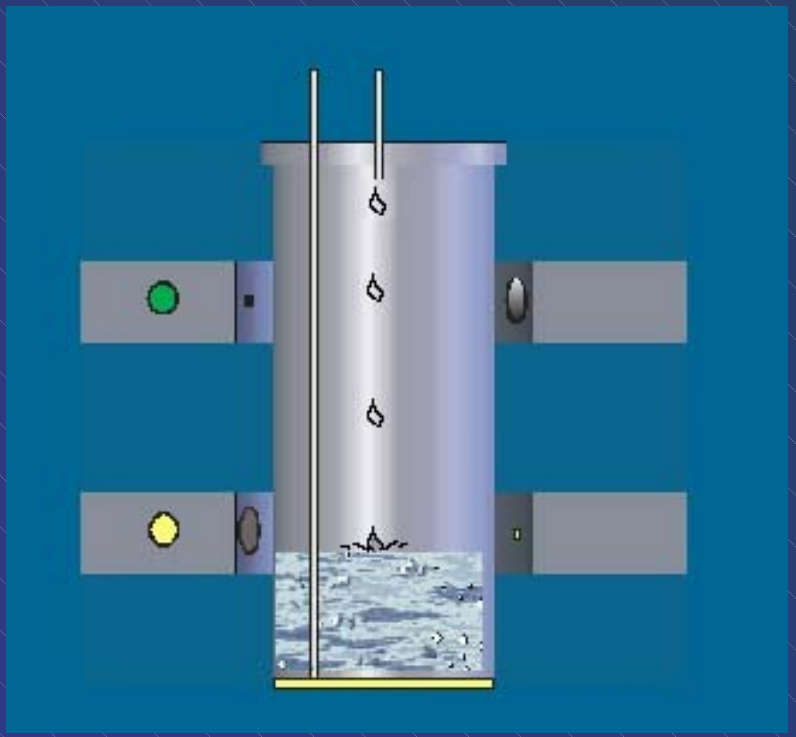
- Second endpoint option: stop evaporation process at lower sensor level (desirable when semivolatile analytes present).
- Level sensor height is adjusted to produce desired endpoint volume.
- Multiple solvent exchanges can be programmed to completely change from column mobile phase to analysis solvent.

Endpoint Time:
Level Sensor 2



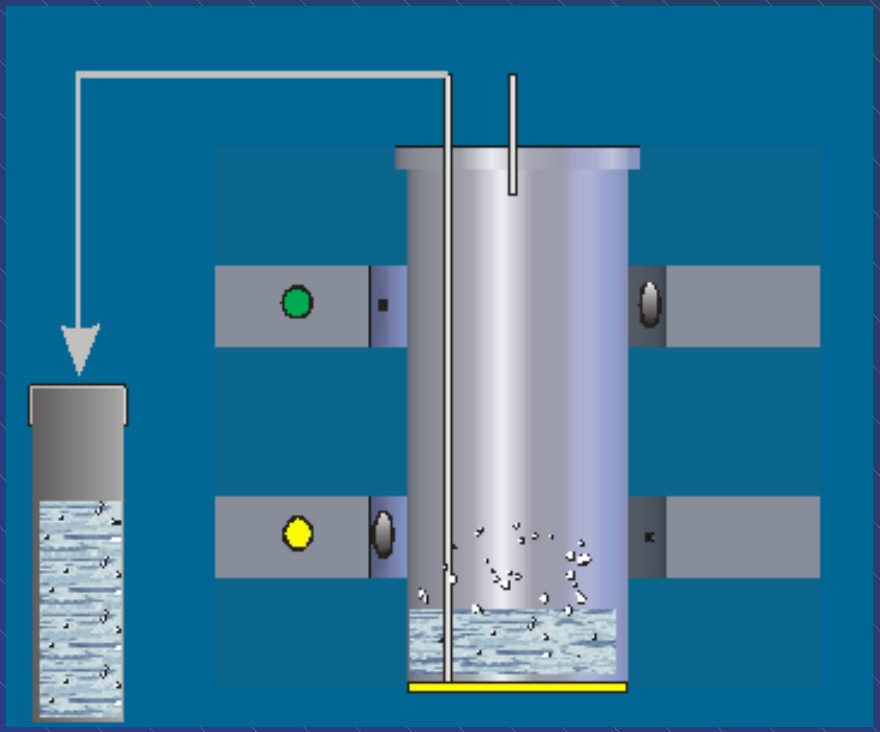
- During solvent exchanges, diluent is added and sample is reconcentrated to lower level sensor using programmed heat and vacuum settings.
- Up to six solvent exchanges can be performed.
- After final solvent exchange sample is evaporated until it reaches lower level sensor.

Fill to level sensor



- Lower level sensor is calibrated to known volume
- After boiling, sample level in chamber is slightly below level of lower sensor
- Diluent slowly added until level sensor is activated
- Transfer out all or an aliquot of sample
- Precision: ± 20µL

Endpoint Time:
Level Sensor 3



- Exchanged sample is transferred to collect vial.
- Programmed rinse solvent added to chamber, heated and mixed with syringe pump, then also transferred to collect vial (ensures quantitative transfer).
- Up to six rinses and transfers can be performed.

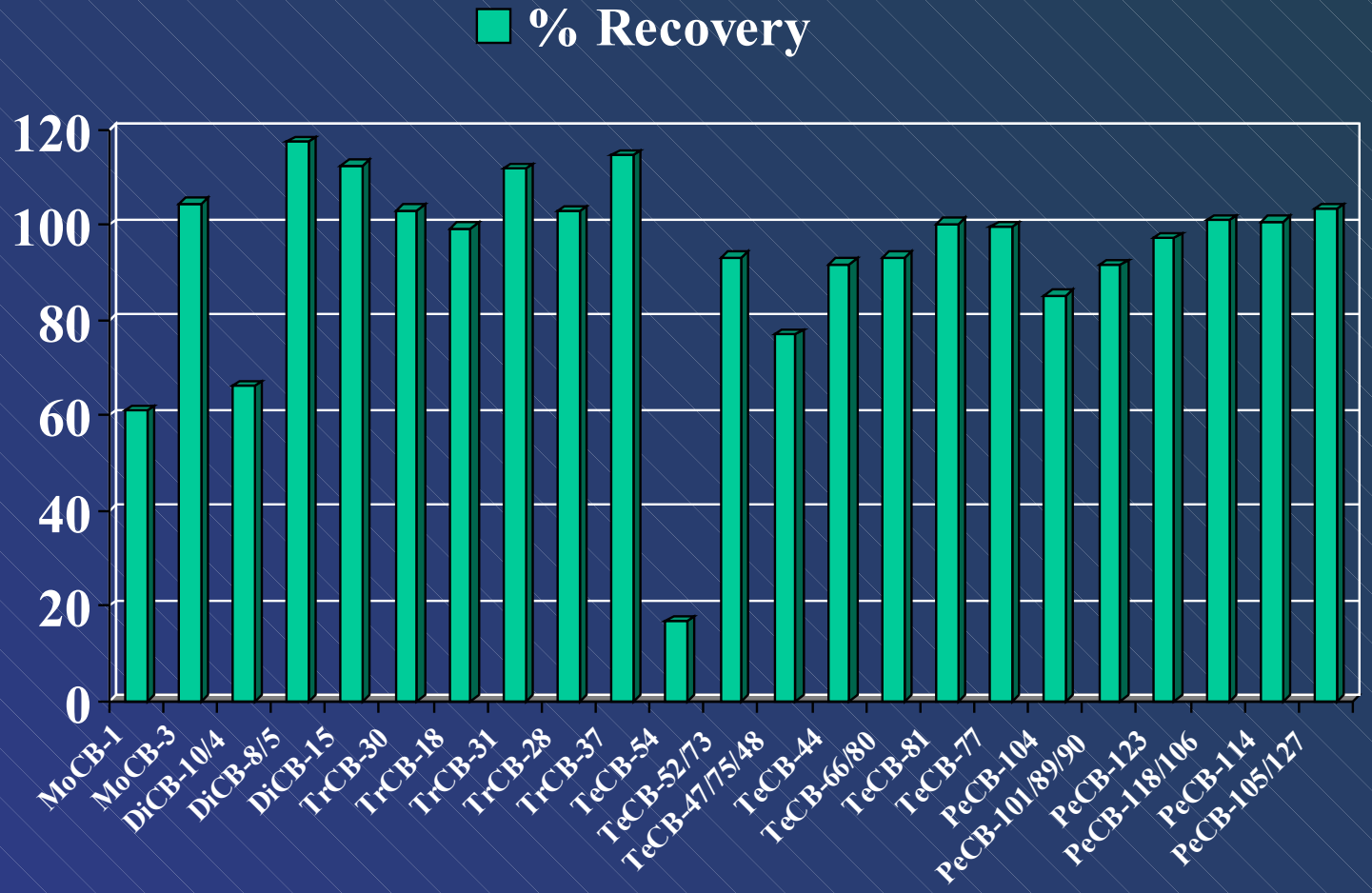
Evaporation Case Studies

- Consistent inter-analyte recoveries
  - *Case 1*: PCBs in environmental samples by isotopic dilution GC/MS
  - *Case 2*: Chlorinated pesticides in red meat by GC/ECD
- Consistent intra-analyte recoveries (precision)
  - *Case 3*: European pesticide Method S19

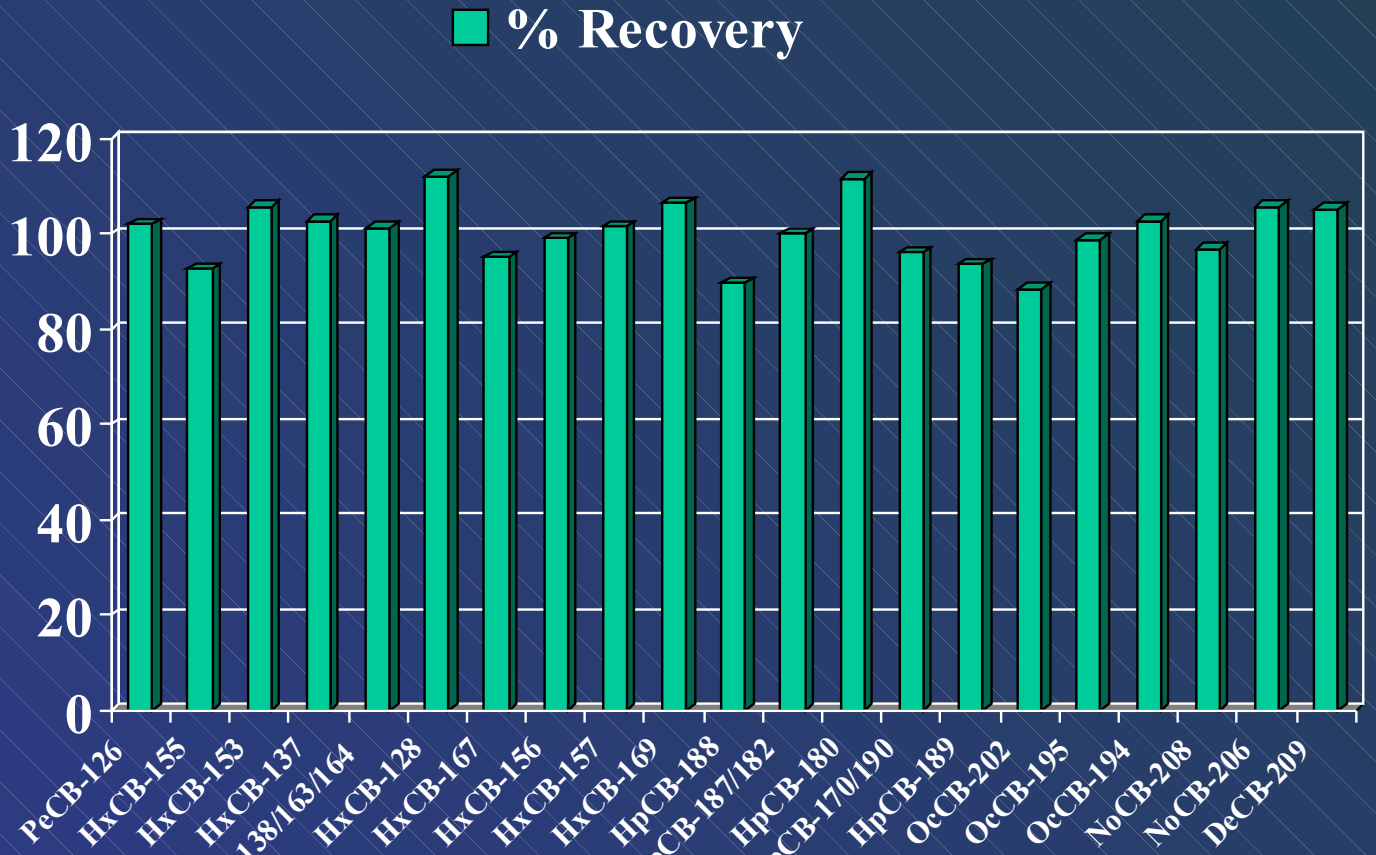
Case #1: PCBs by GC/MS

- Soxhlet extraction of spiked thimble (LCS)
  - 300 mL toluene, reflux 16 hours
- Concentration and exchange to hexane (100 mL)
  - a) rotary evaporation, 65 °C, 25# vacuum, ***or***
  - b) automated GPC + on-line evaporation (AccuVap)
- Cleanups as needed
- Add IS, concentrate to 20 µL with N_2

PCBs w/ Rotary Evaporation



PCBs w/ Rotary Evaporation



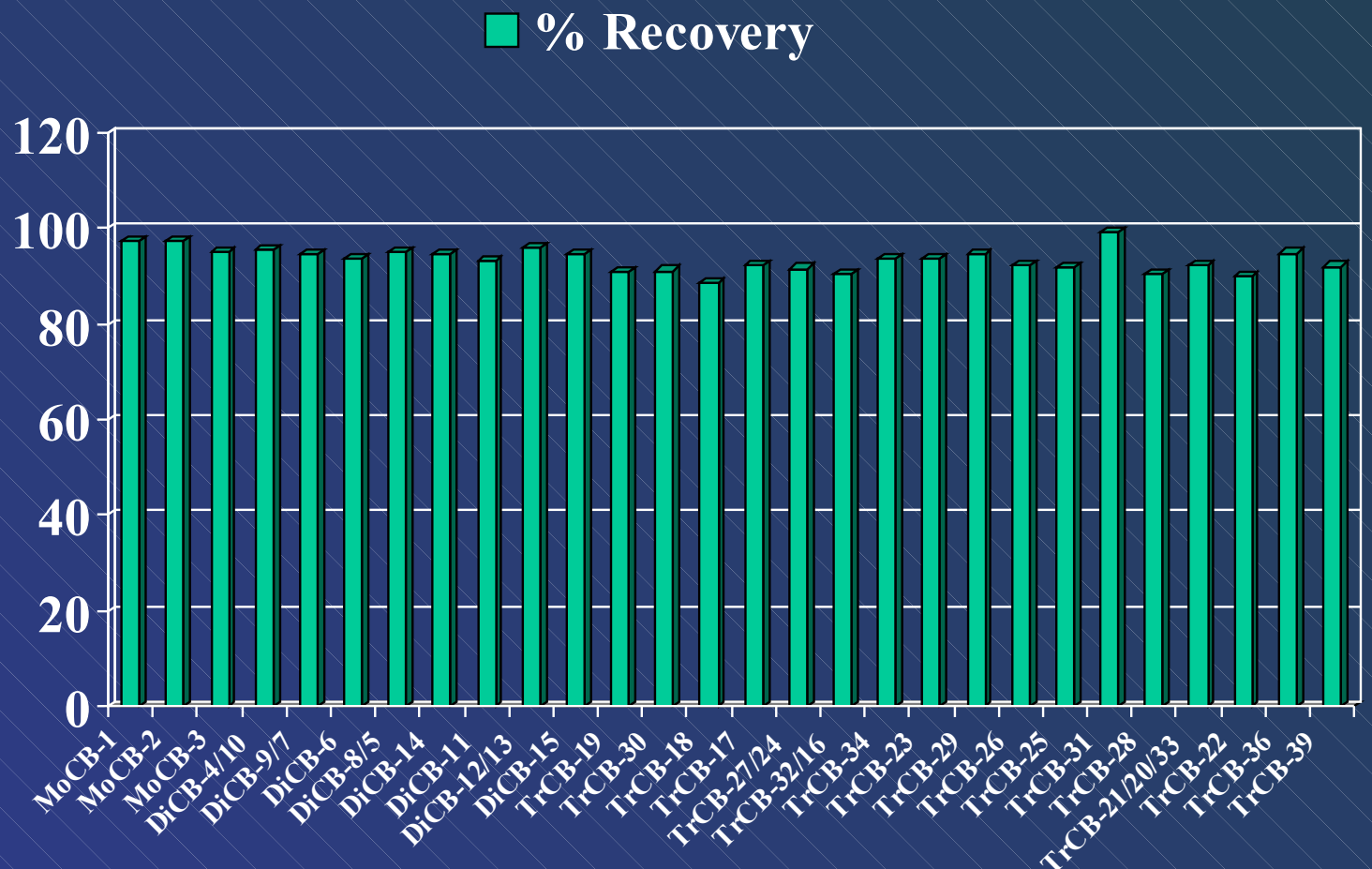
Automated Evaporation

Case 1: PCBs by GC/MS

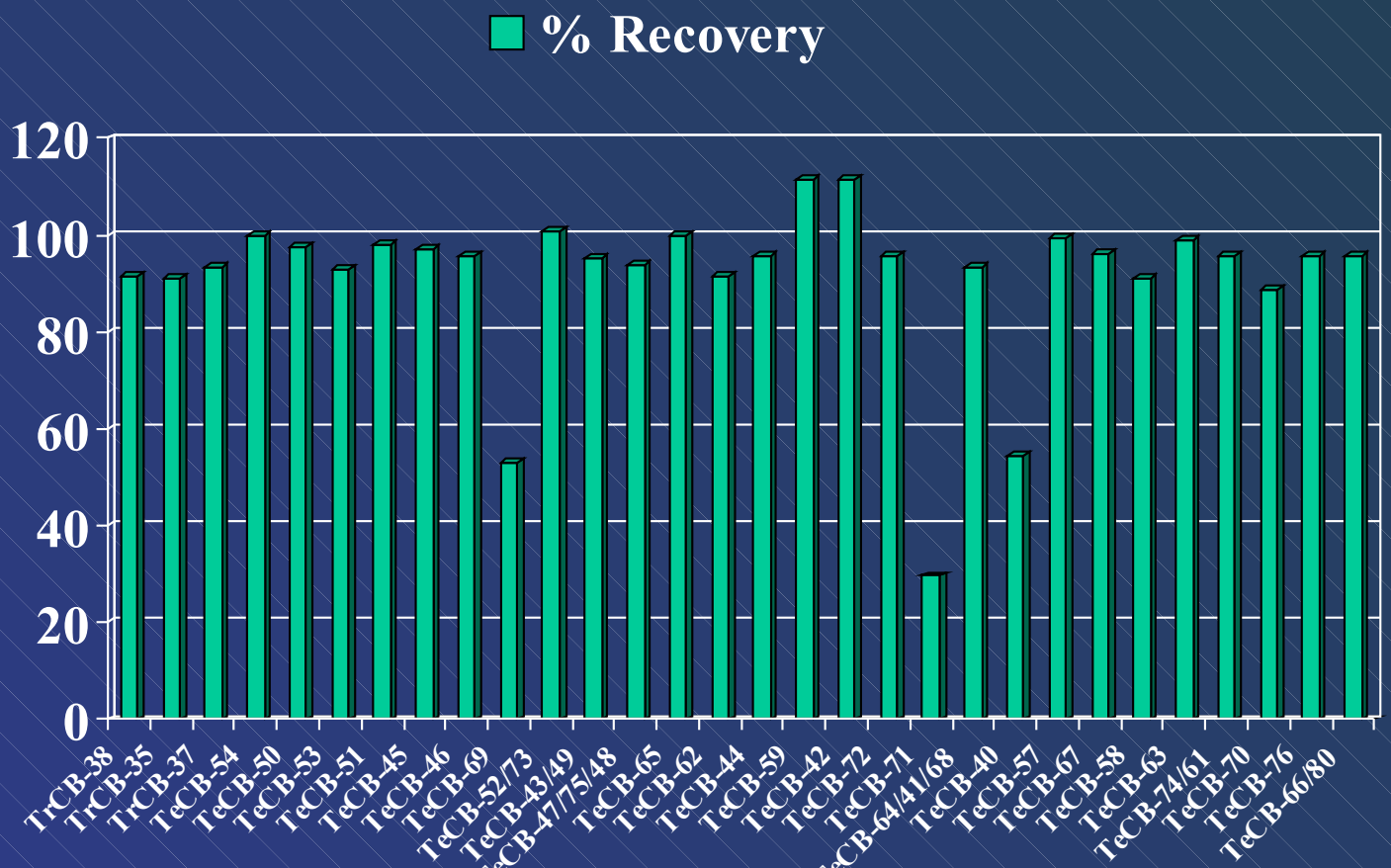
Endpoint: dryness
Keeper: none
Diluent transfer volume: 2.5 mL
Transfers: 1
Final solvent: hexane

	Temp (°C)	Press (Torr)
Zone 1	15	300
Zone 2	28.5	300
Zone 3	31.5	300
Endpoint	32	300

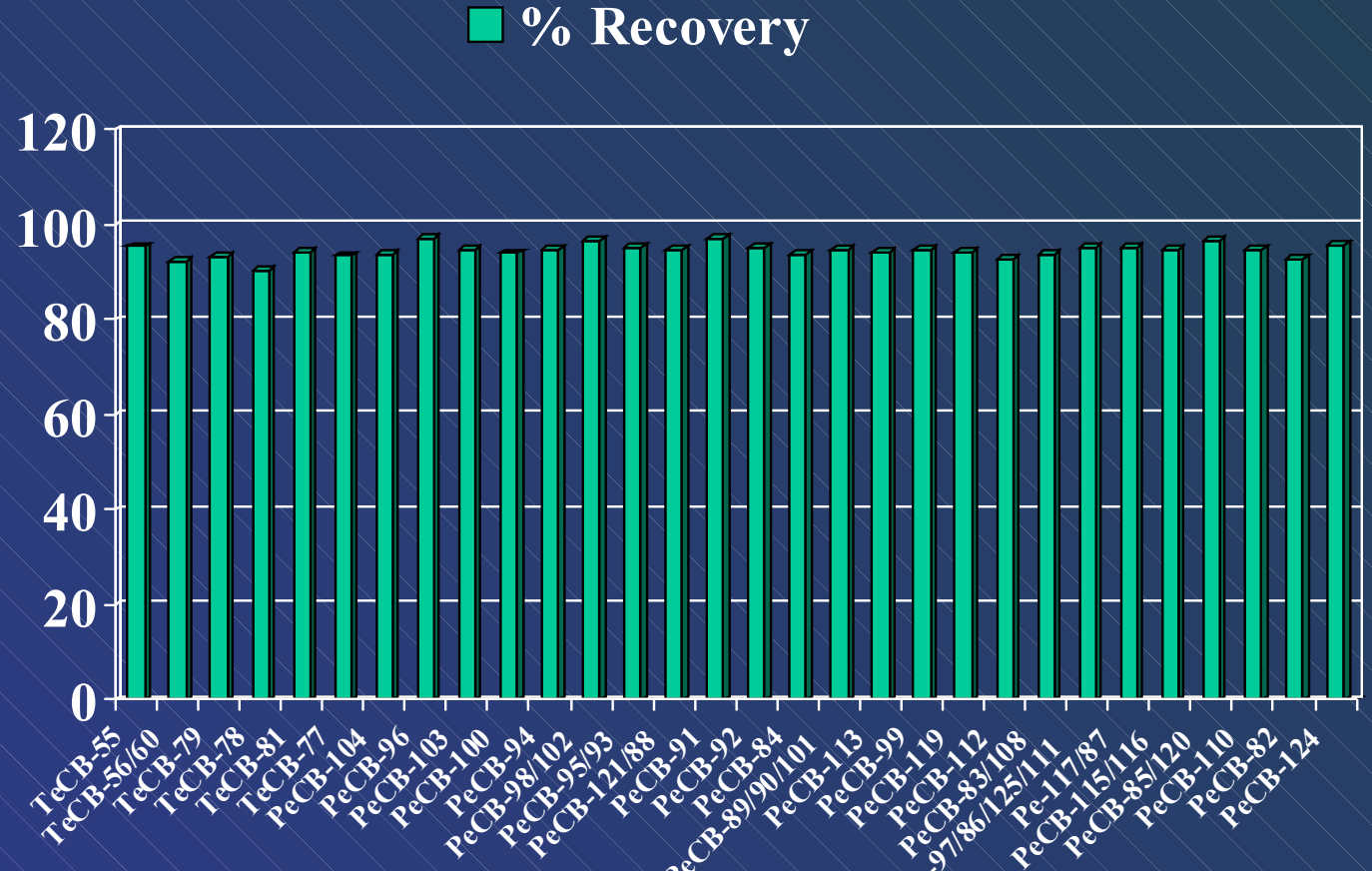
PCB with automated evaporation



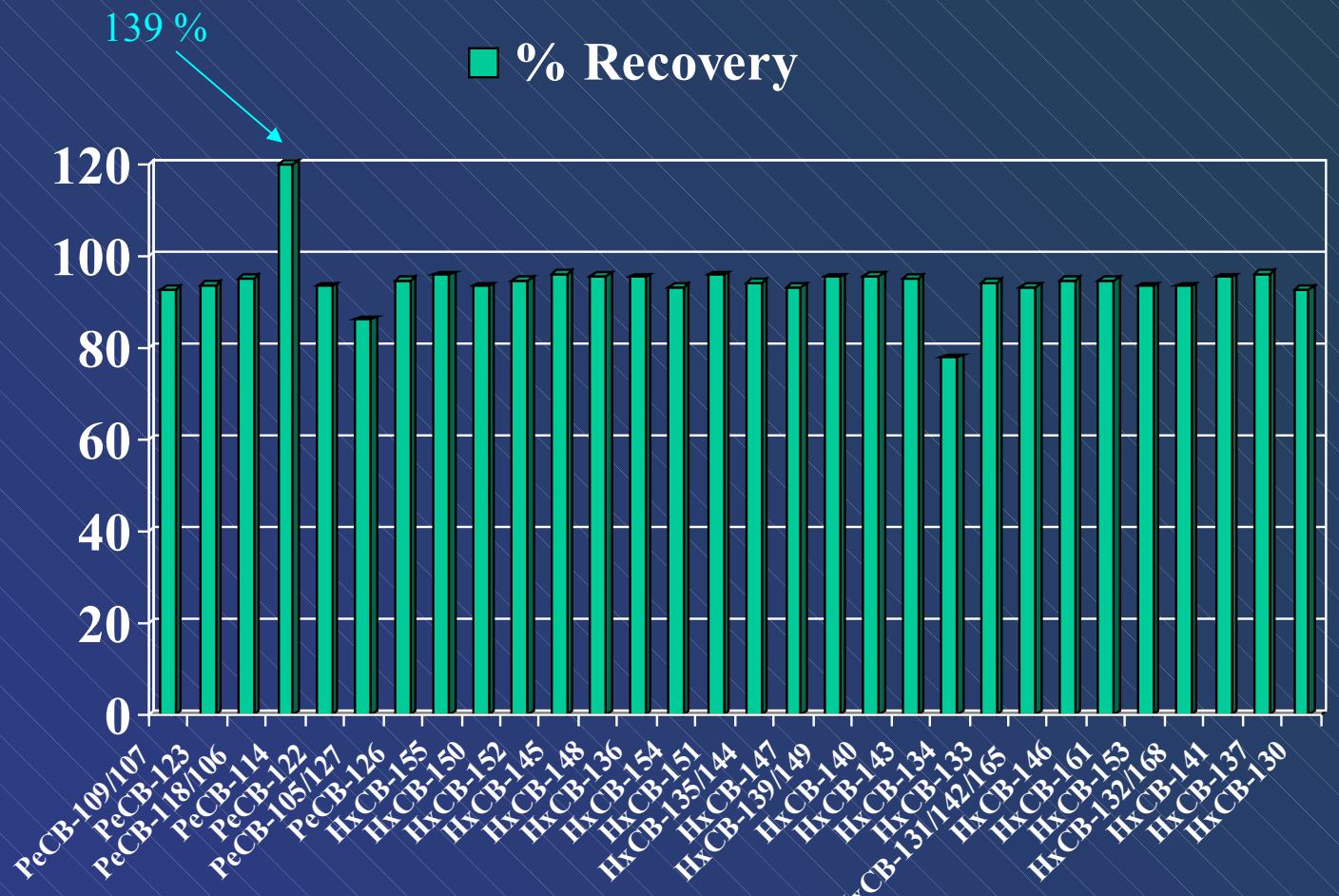
PCB with automated evaporation



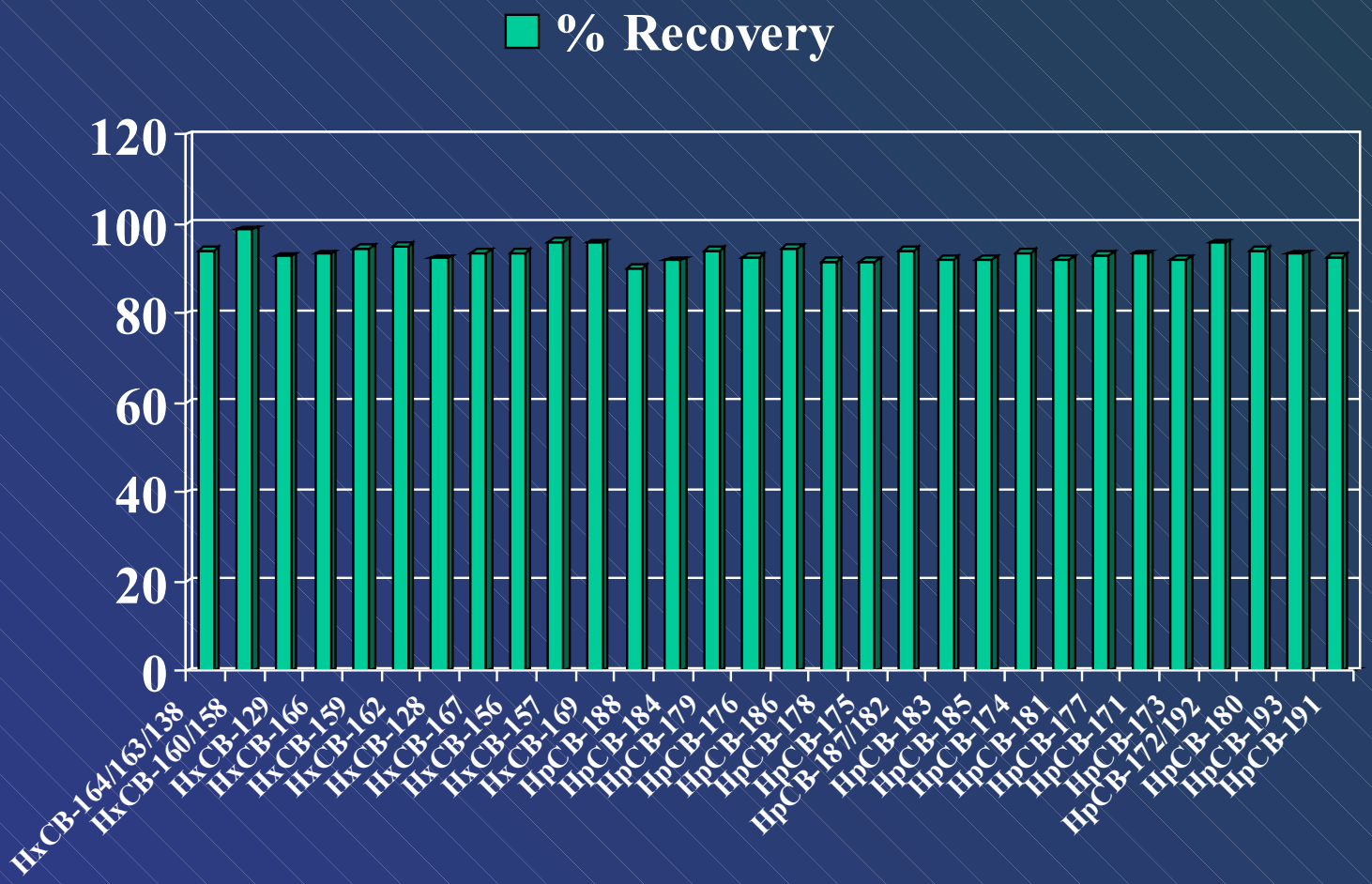
PCB with automated evaporation



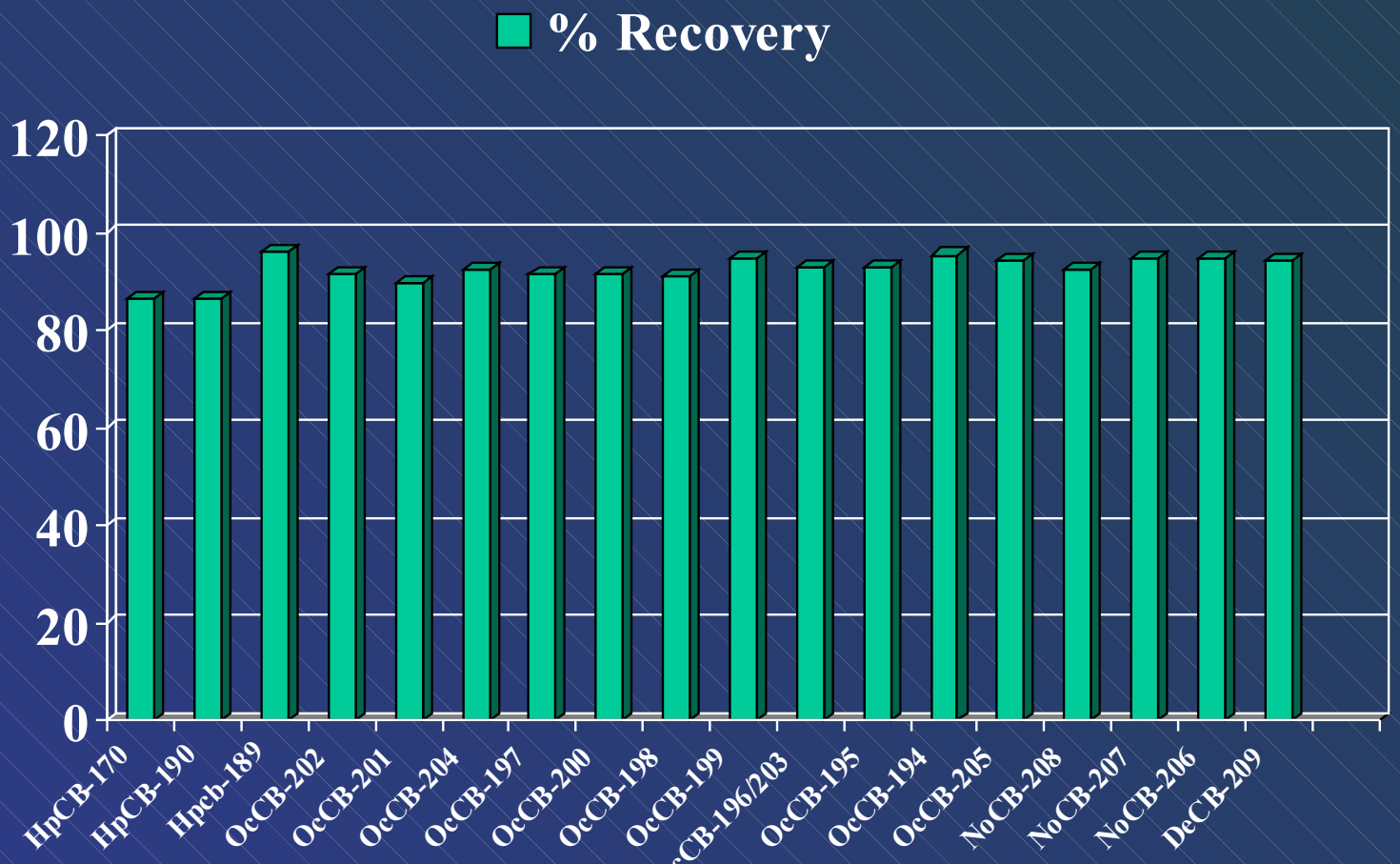
PCB with automated evaporation



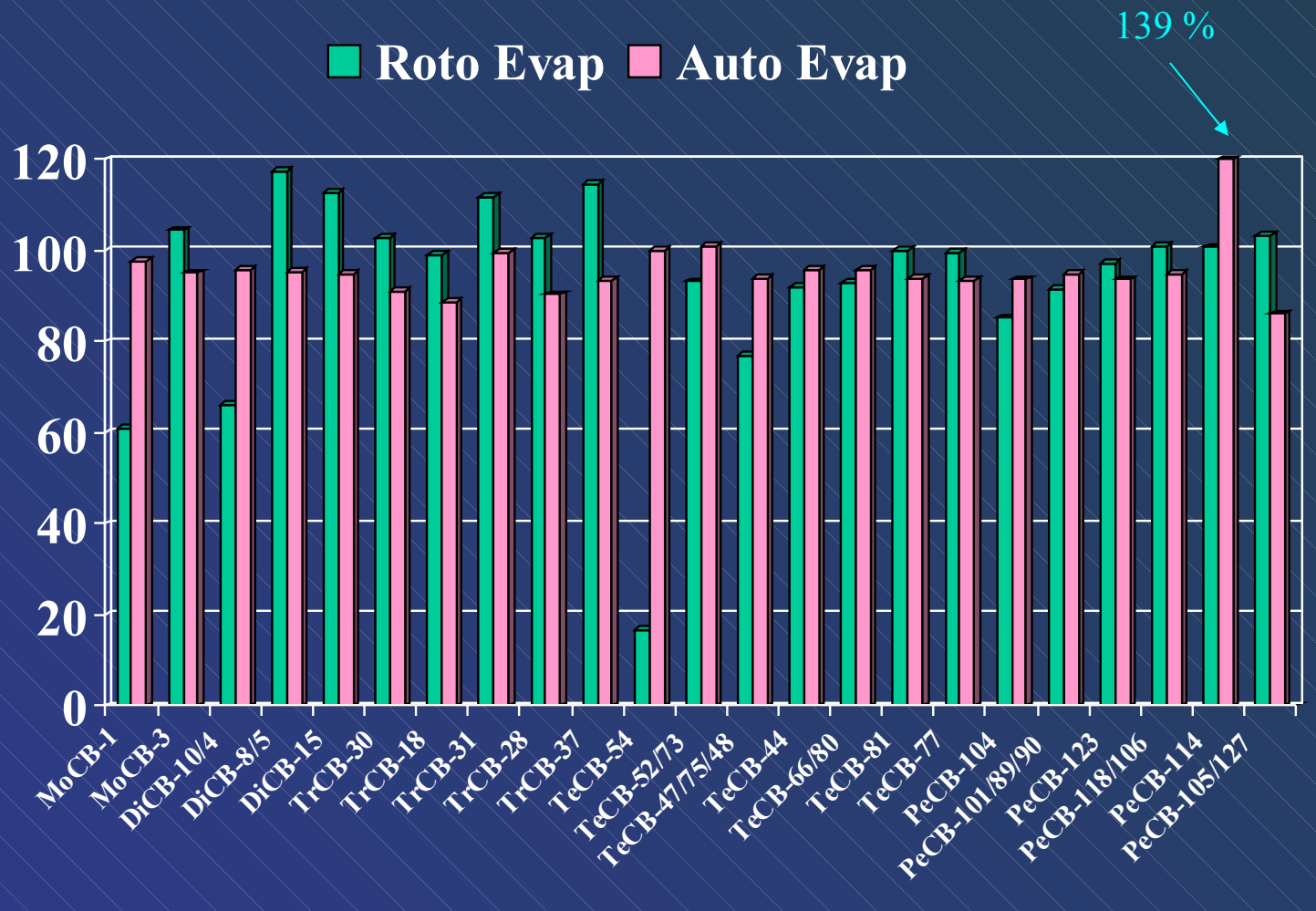
PCB with automated evaporation



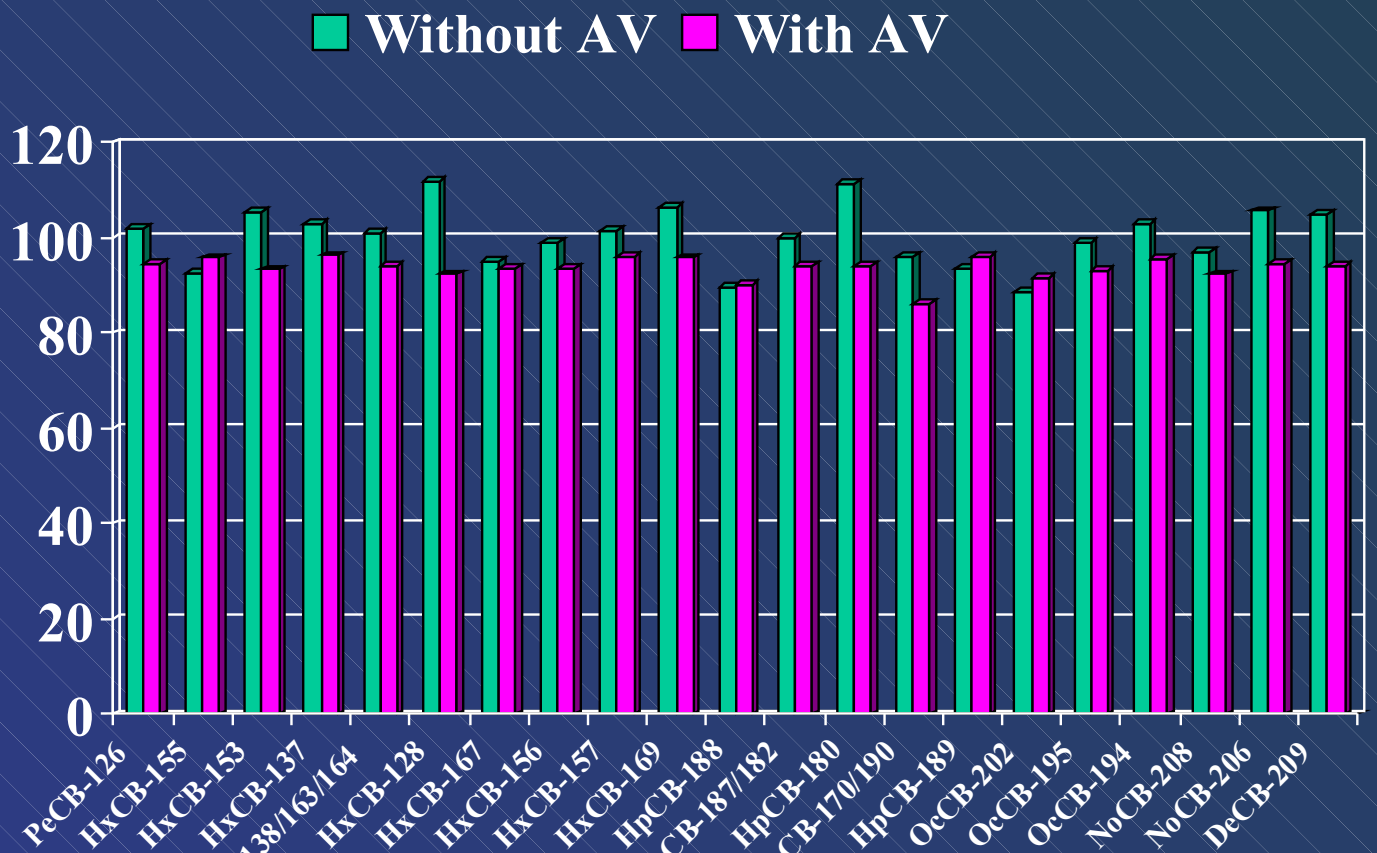
PCB with automated evaporation



Compare PCB Congeners-1



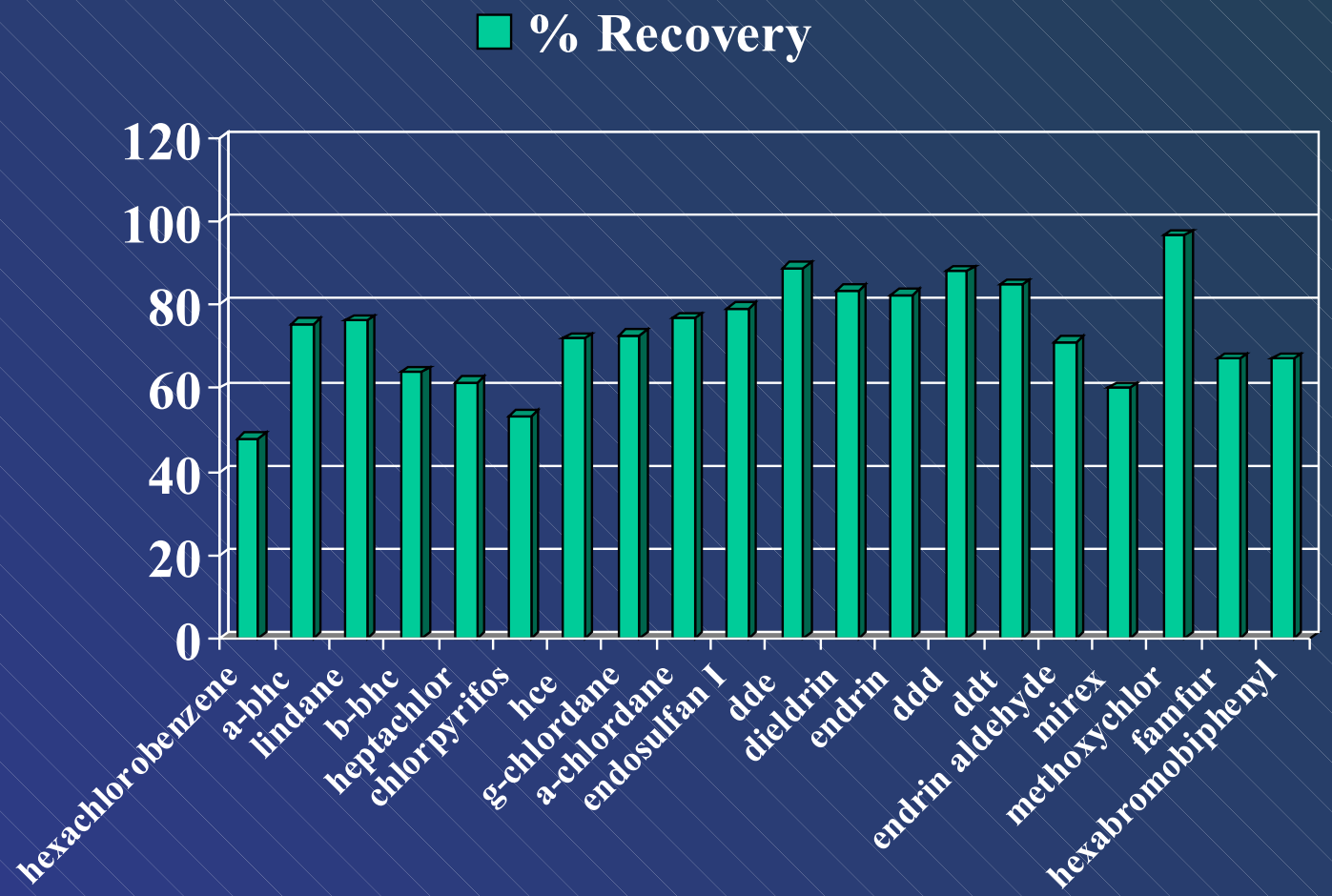
Compare PCB Congeners-2



Case #2: Pesticides in Red Meat

- Rendered fat sample
  - GPC with 100% DCM
    - Rotovap @ 45 °C, or
    - Automated evaporation (AccuVap)
  - GC/ECD

Case 2: Rotary Evaporation



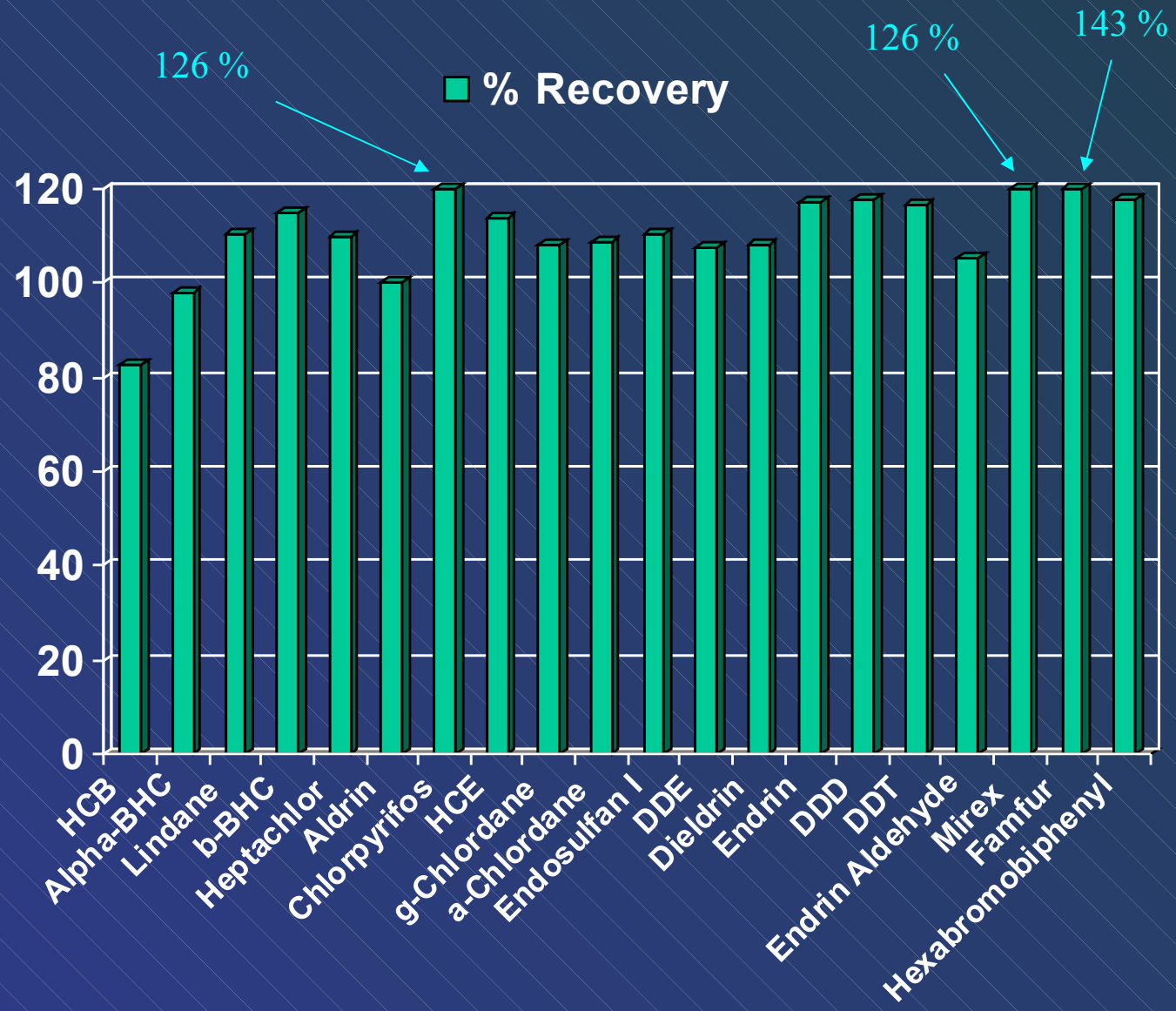
Automated Evaporation

Case 2: chlorinated pesticides in meat

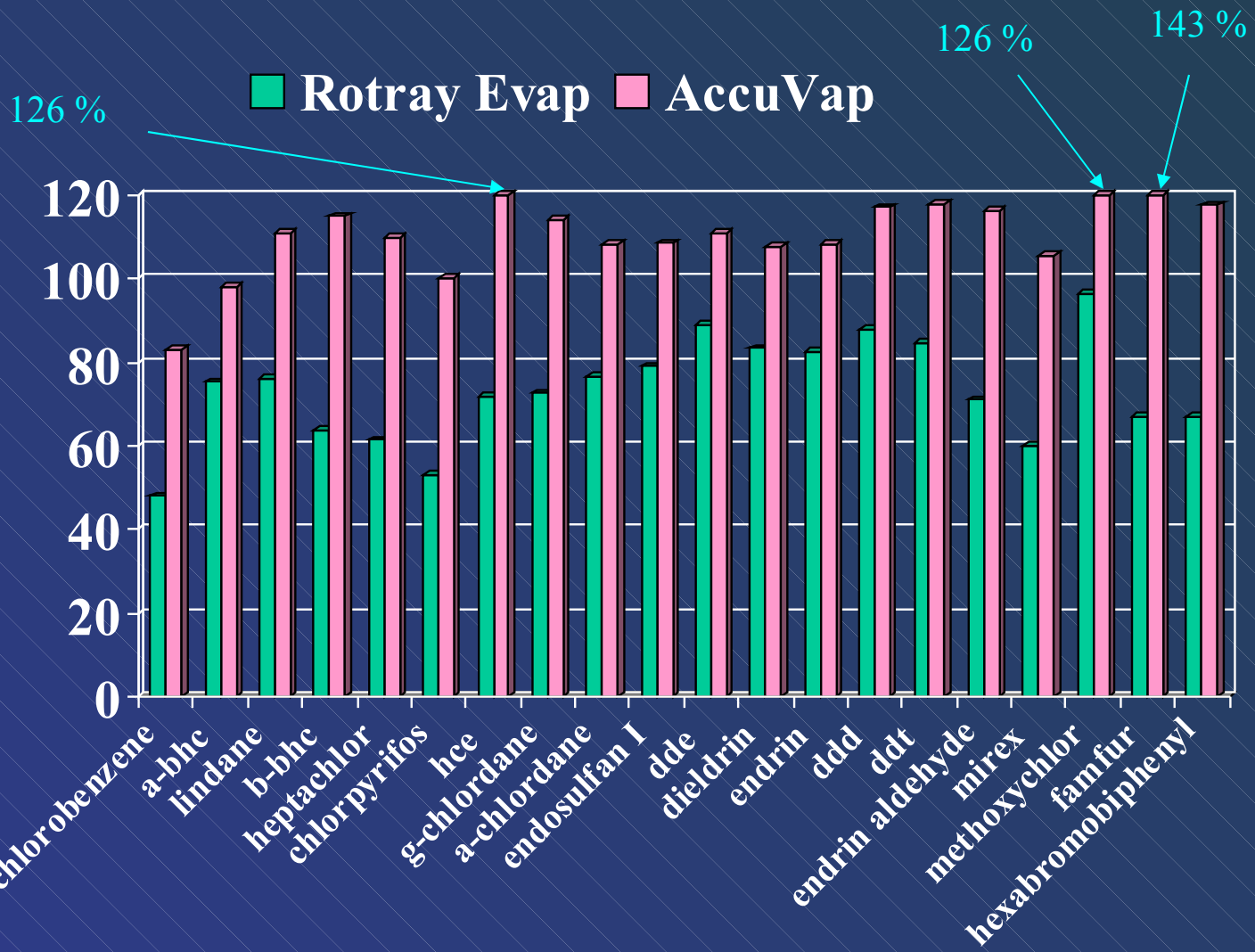
Endpoint: dryness
Keeper: none
Diluent transfer volume: 5 mL
Transfers: 1
Final solvent: hexane

	Temp (°C)	Press (Torr)
Zone 1	18	300
Zone 2	30	300
Zone 3	35	300
Endpoint	35	300

Case 2: AccuVap Recoveries



Compare Case #2



Case #3: European GC/MS Method

(S19 multiresidue pesticide method)

- Spiked blanks (LCS)
- GPC
  - 55g S-X3, 1:1 EtOAc/cyclohexane
  - 5 mL/min, collect from 17 to 37 minutes
  - On-line evaporation with AccuVap
- Fused silica capillary column HP/5/MS
  - 0.25 cm x 30 meter, 0.25 µm film thickness
  - KAS 4 injector (Gerstel), 80 °C
  - 60 °C, 1 min, 10 °C/min to 180 °C, hold 5 min
  - 5 °C/min to 290 °C, hold 10 min
- Agilent 6890 GCMS detector HP 5973

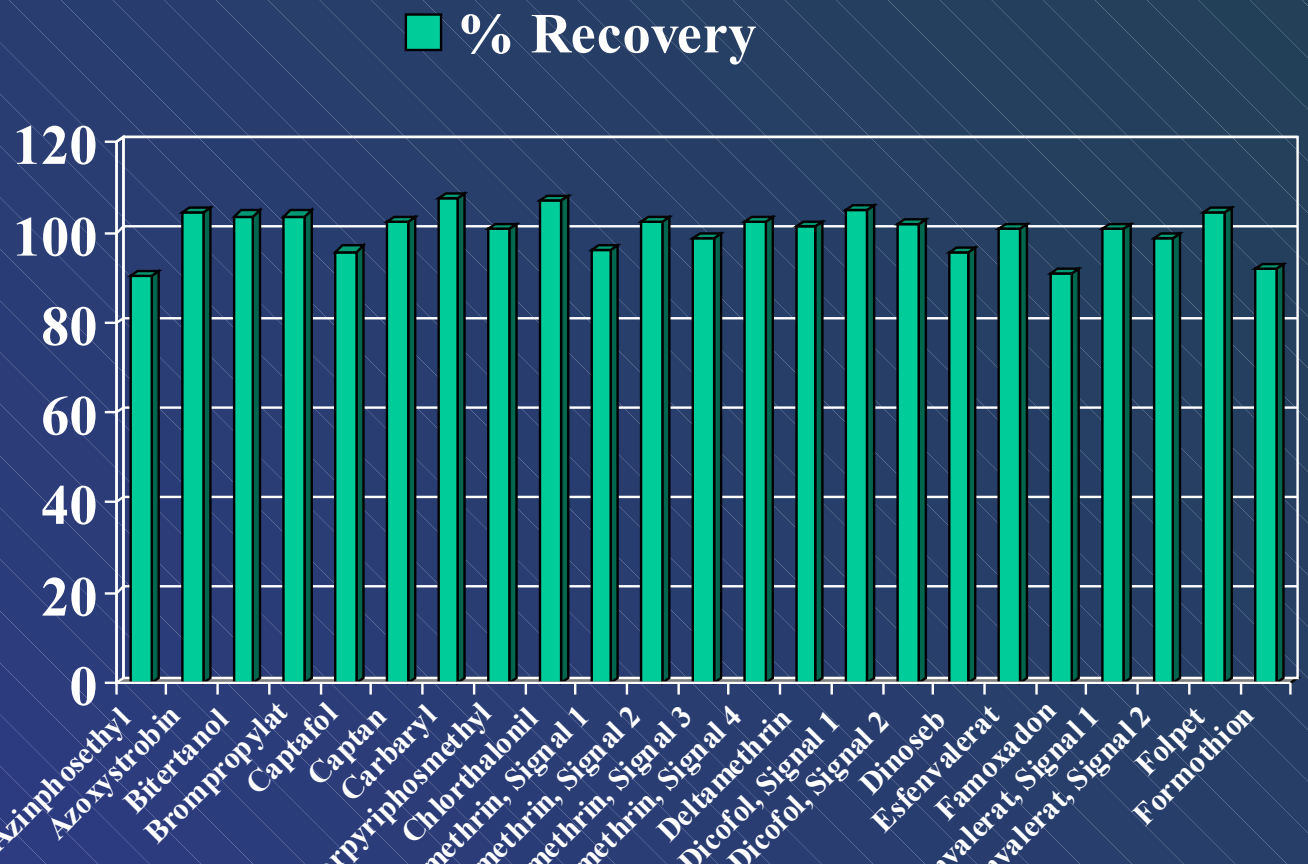
Automated Evaporation

Case 3: S19 multiresidue GC/MS

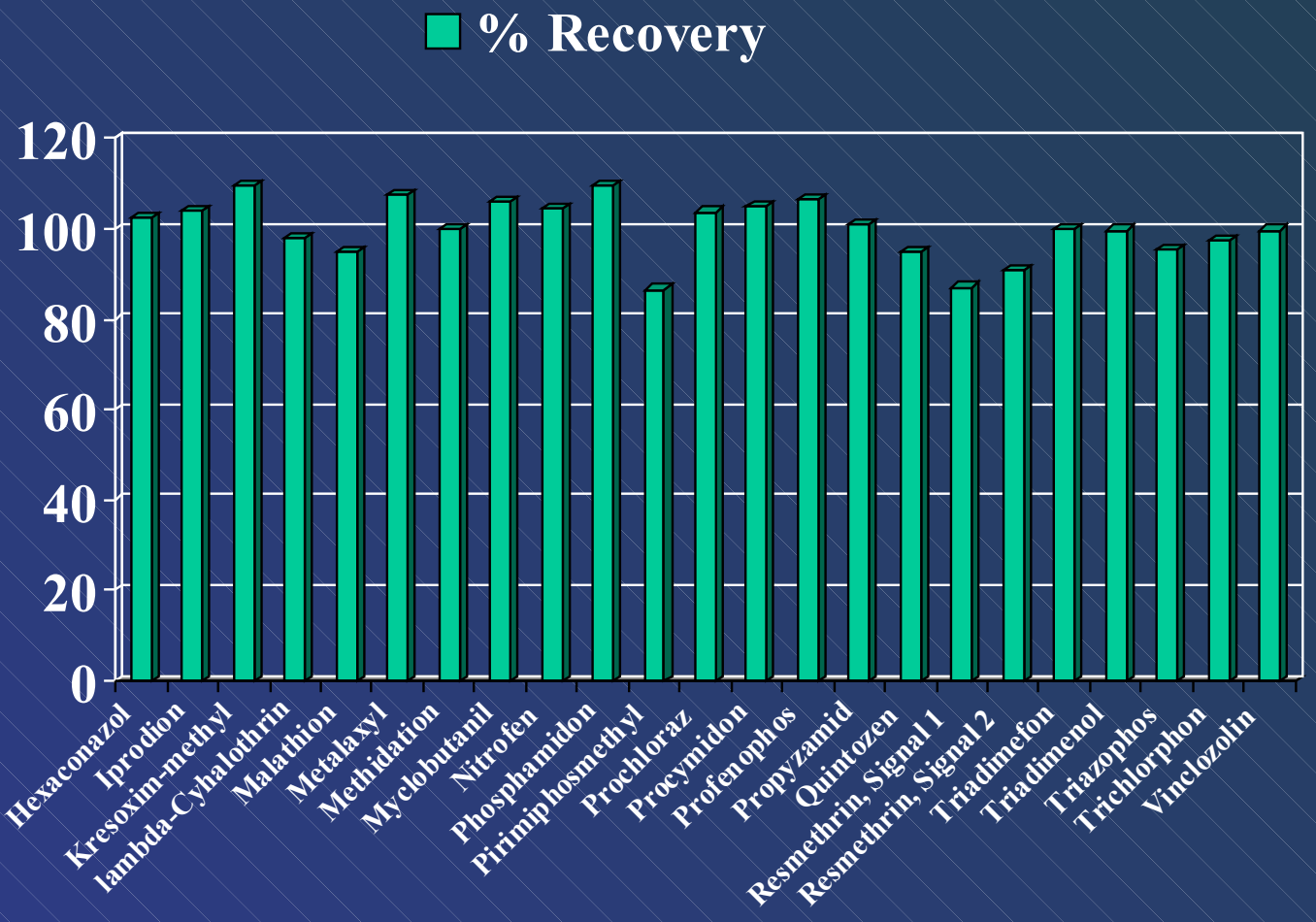
Endpoint: dryness
Keeper: none
Diluent transfer volume: 500 µL
Transfers: 2
Final solvent: cyclohexane

	Temp (°C)	Press (Torr)
Zone 1	30	200
Zone 2	58	200
Zone 3	64	200
Endpoint	65	200

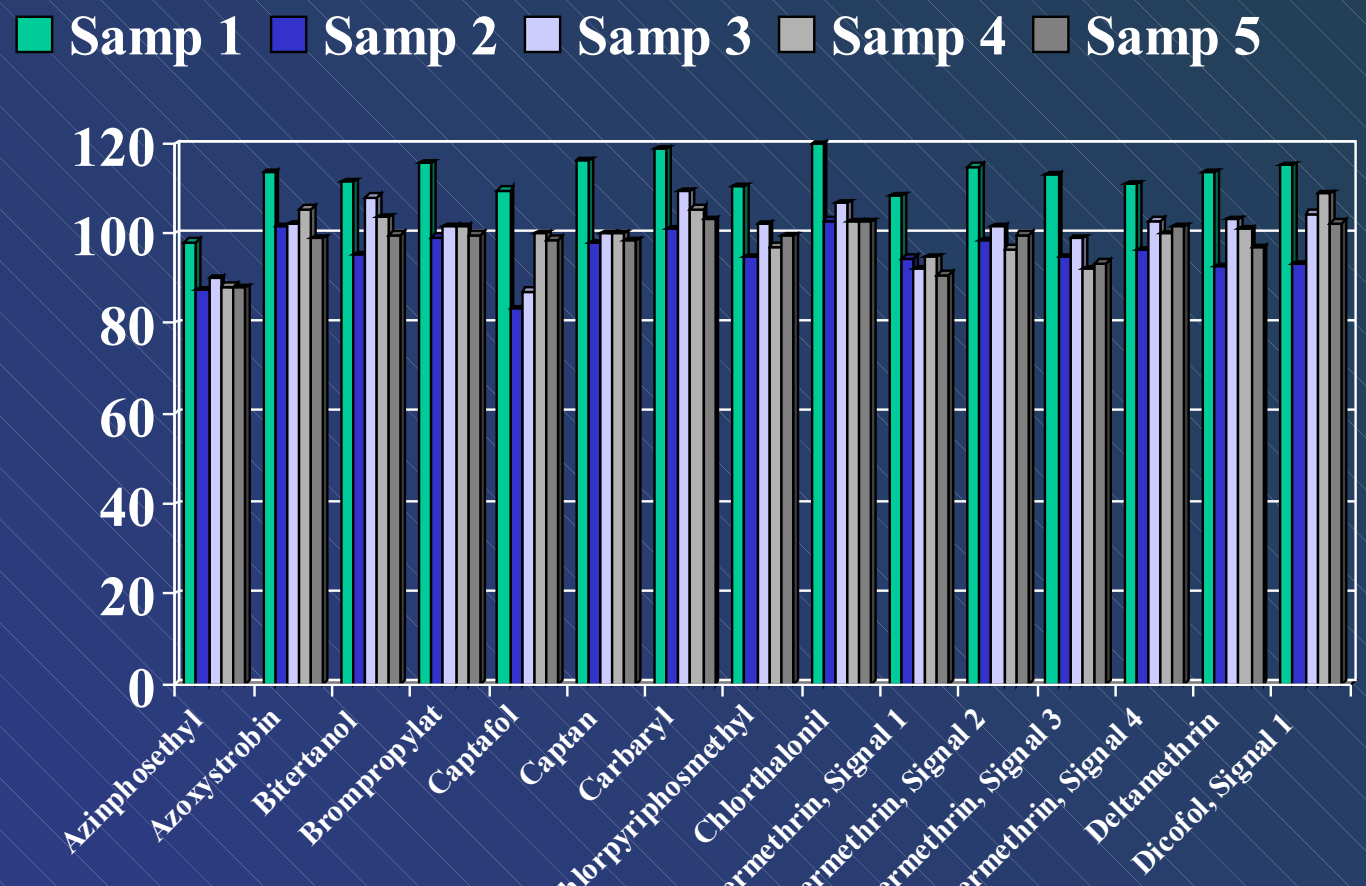
S19 Summary PSM - 1



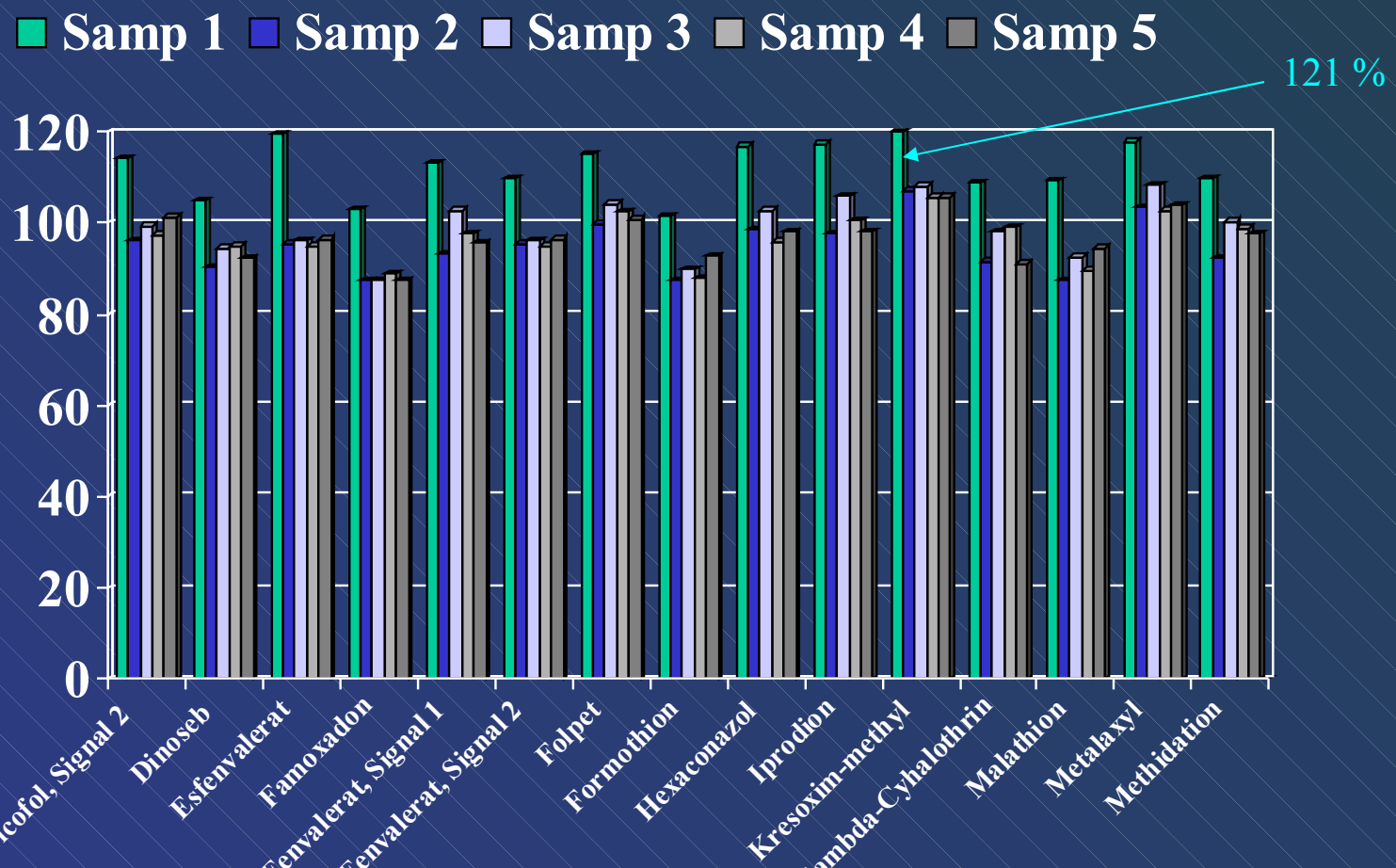
S19 Summary PSM - 2



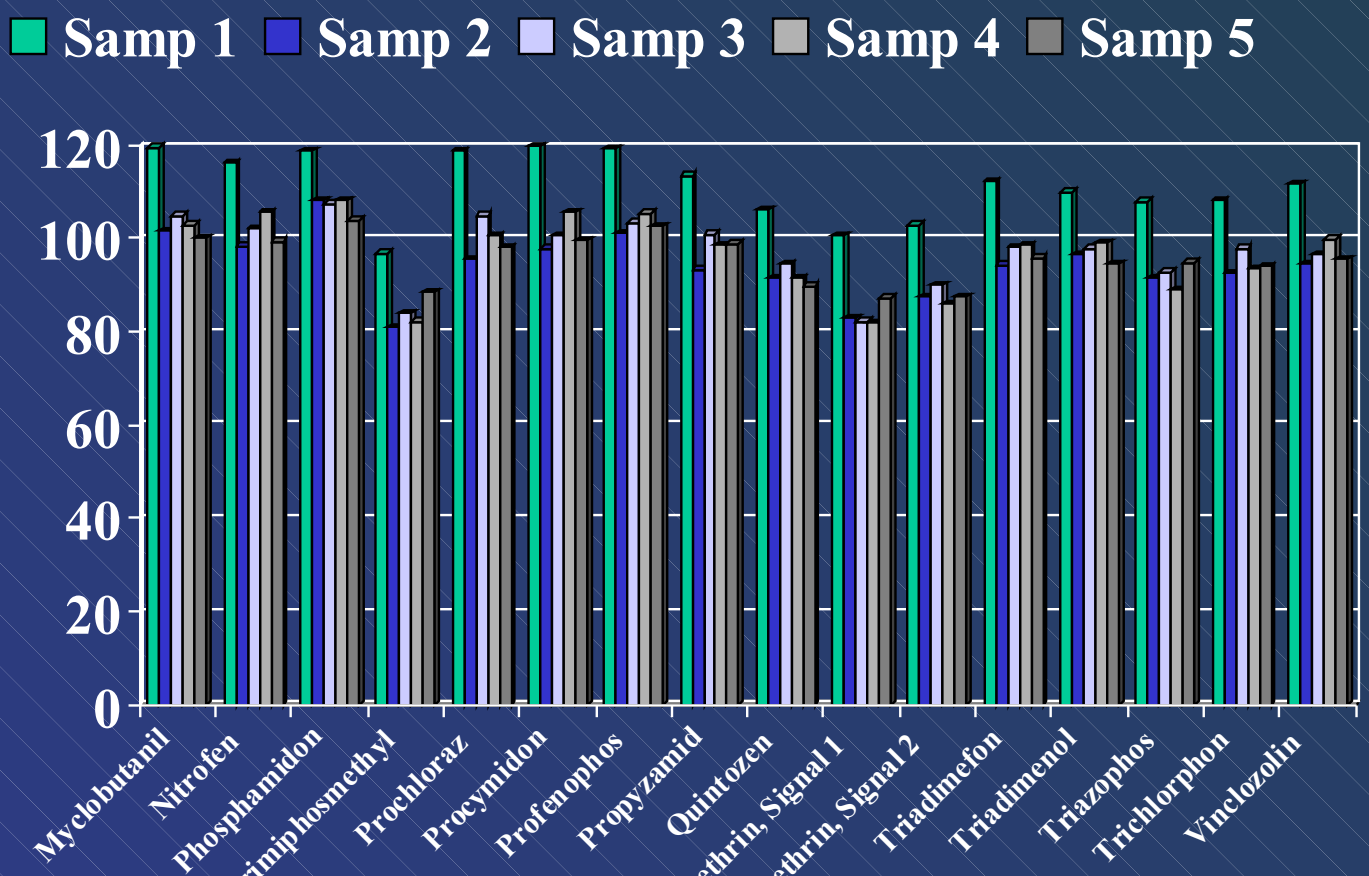
S19 Replicates PSM - 1



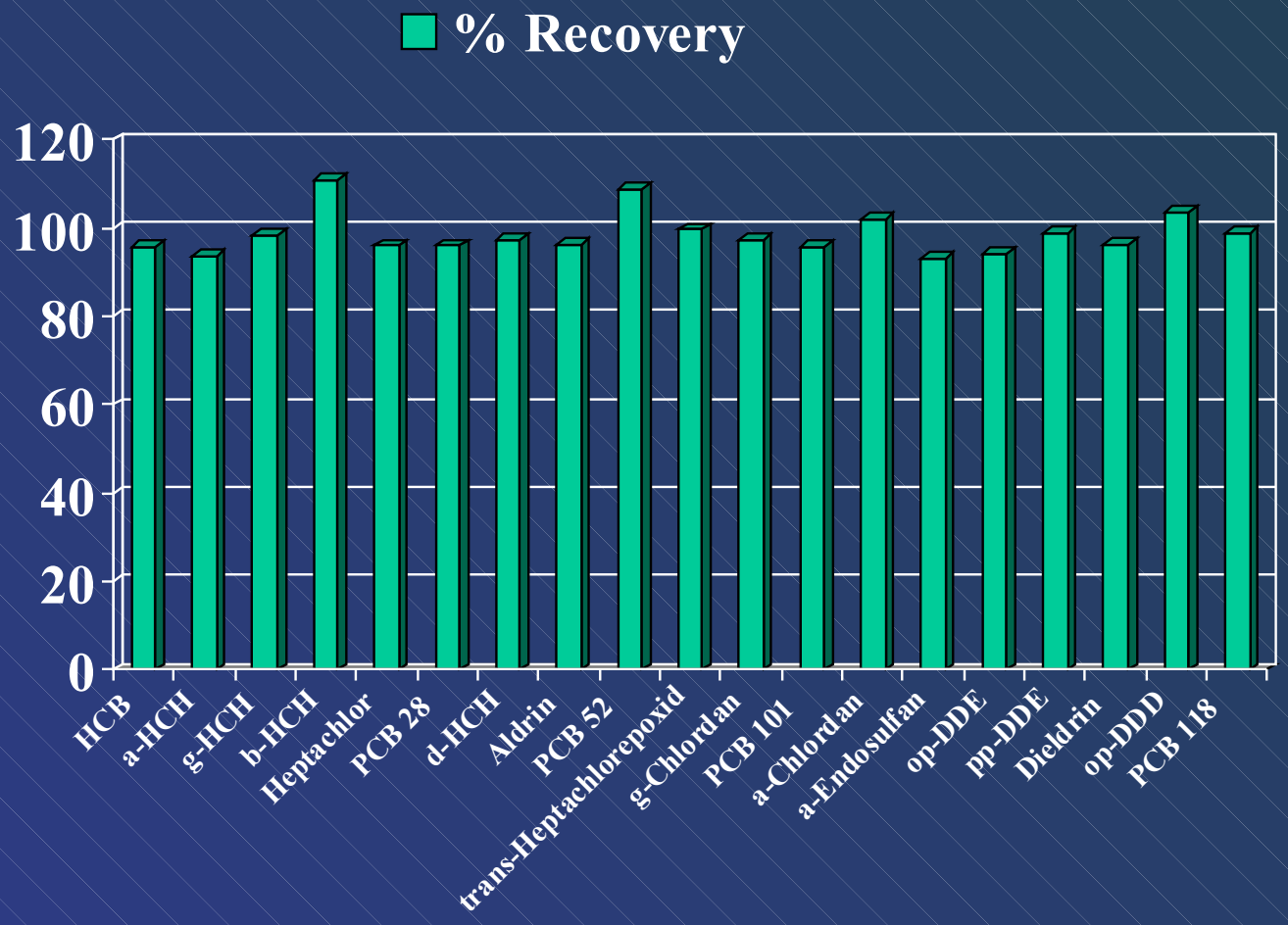
S19 Replicates PSM - 2



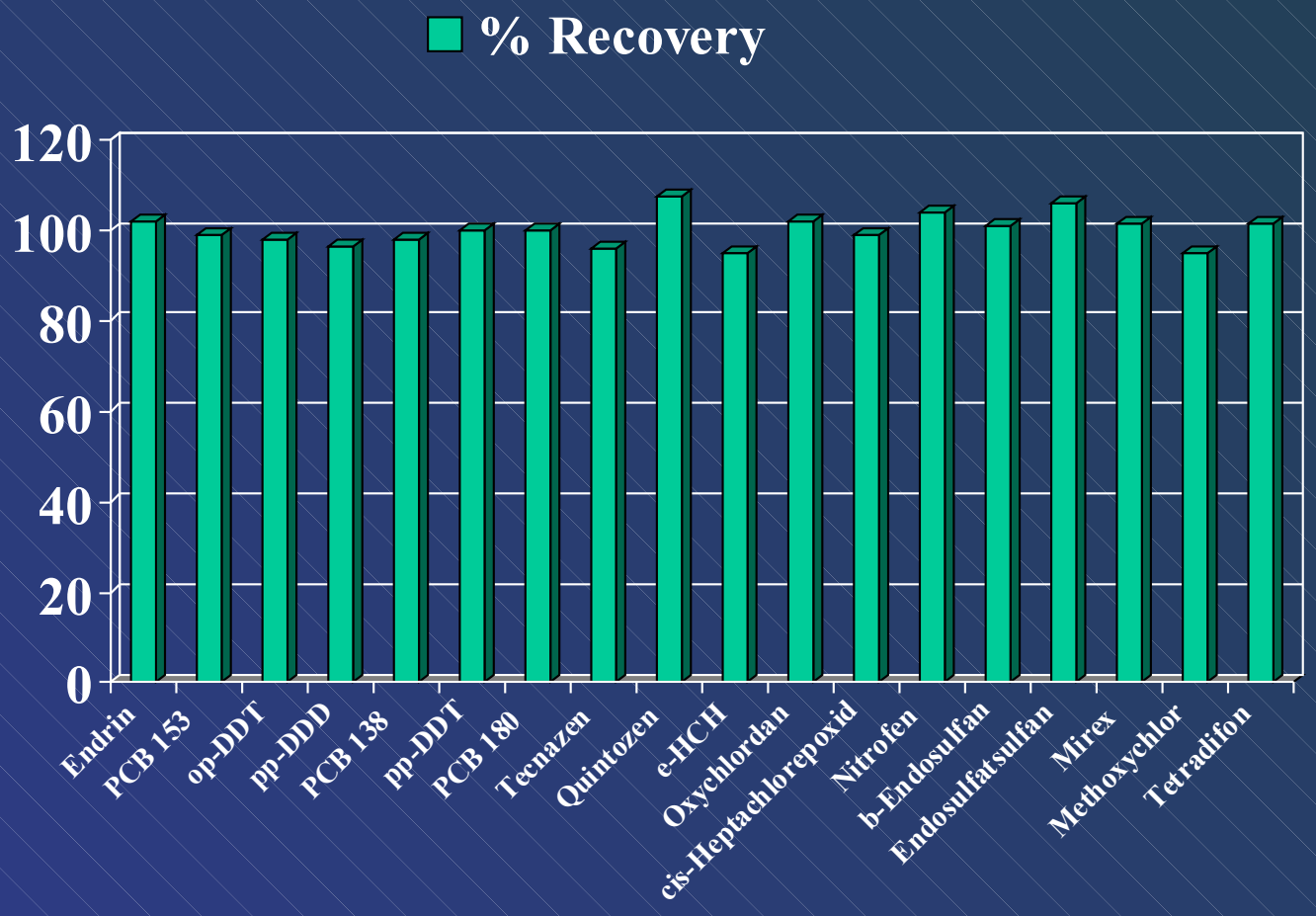
S19 Replicates PSM - 3



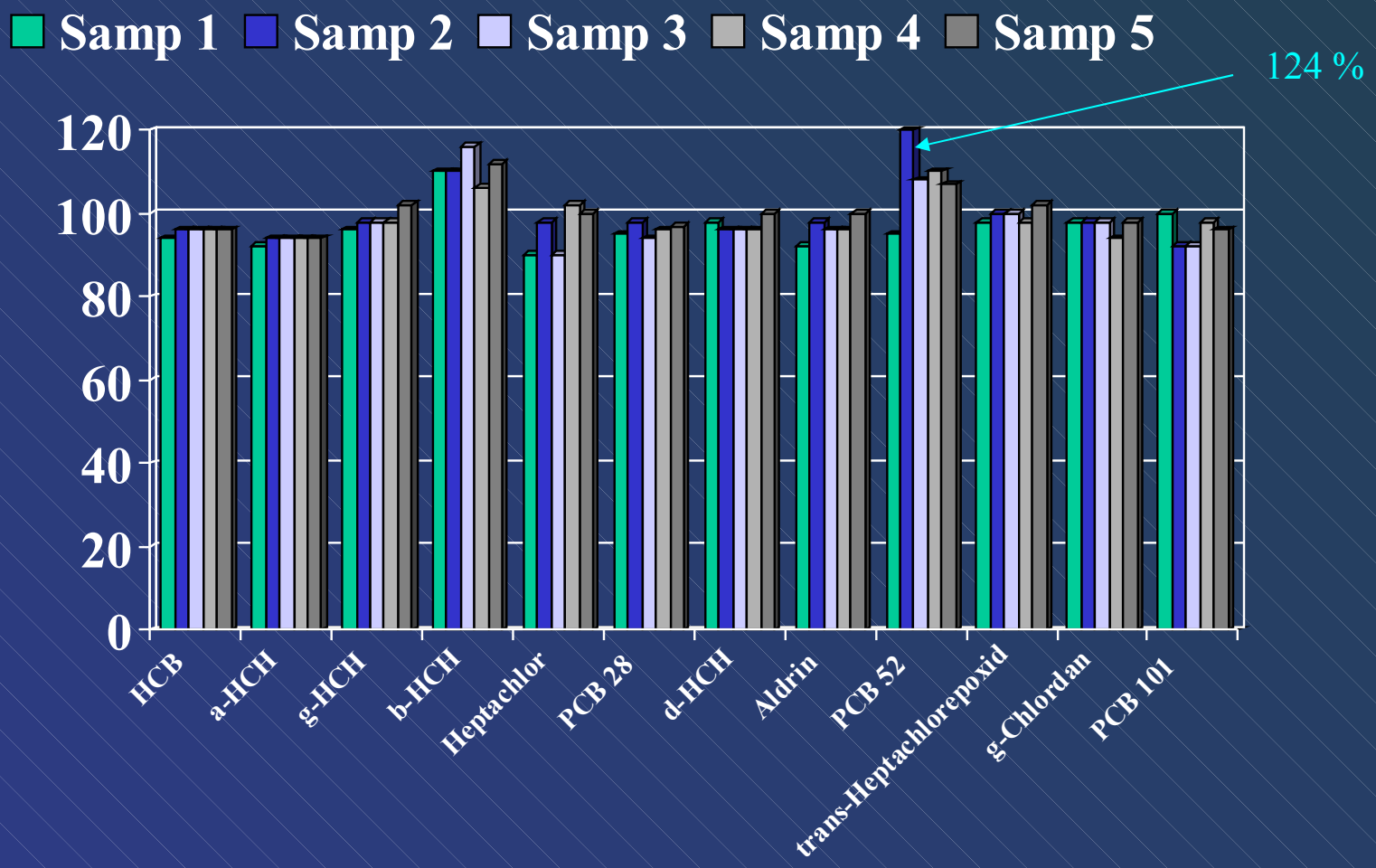
S19 Summary OCP - 1



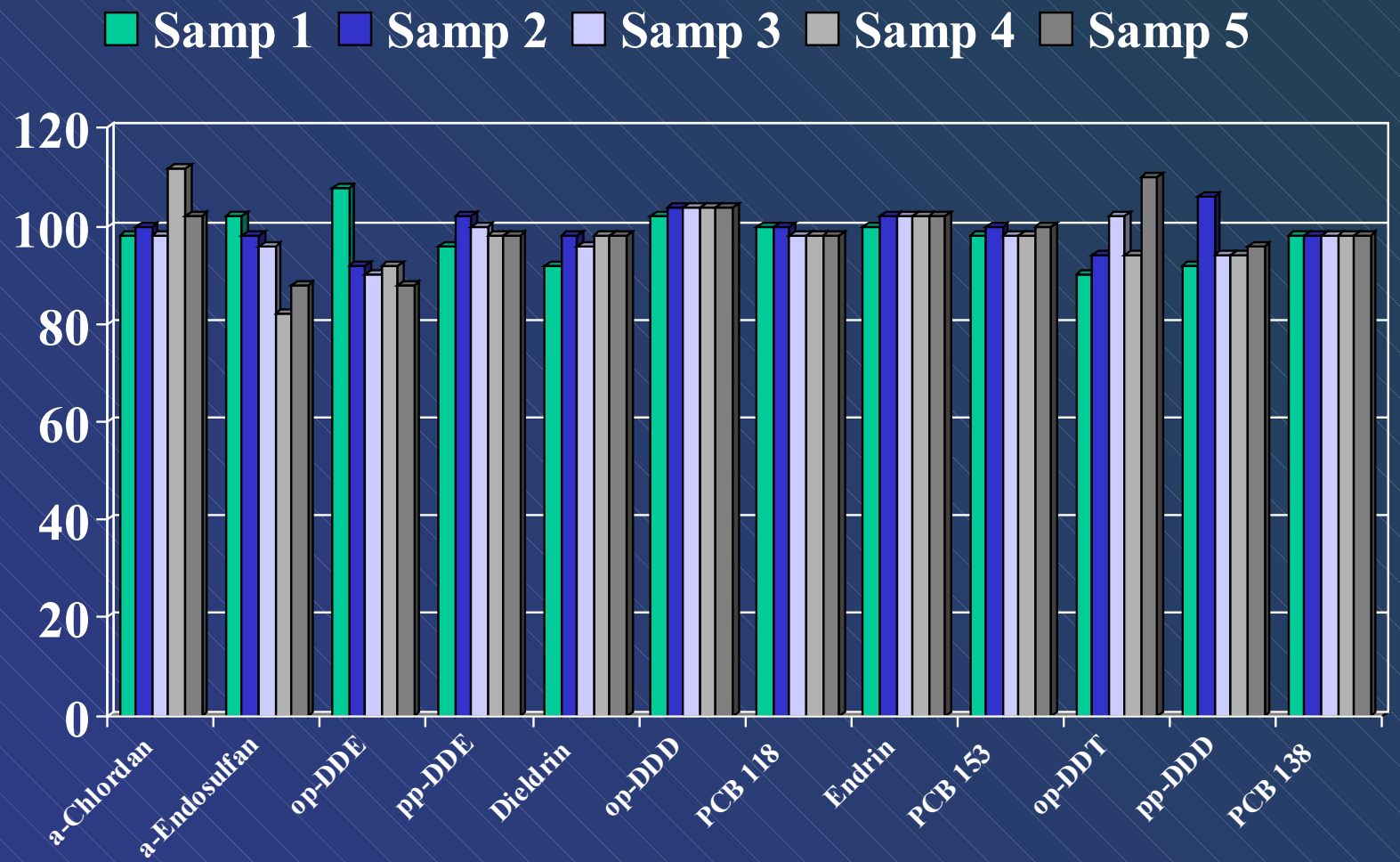
S19 Summary OCP - 2



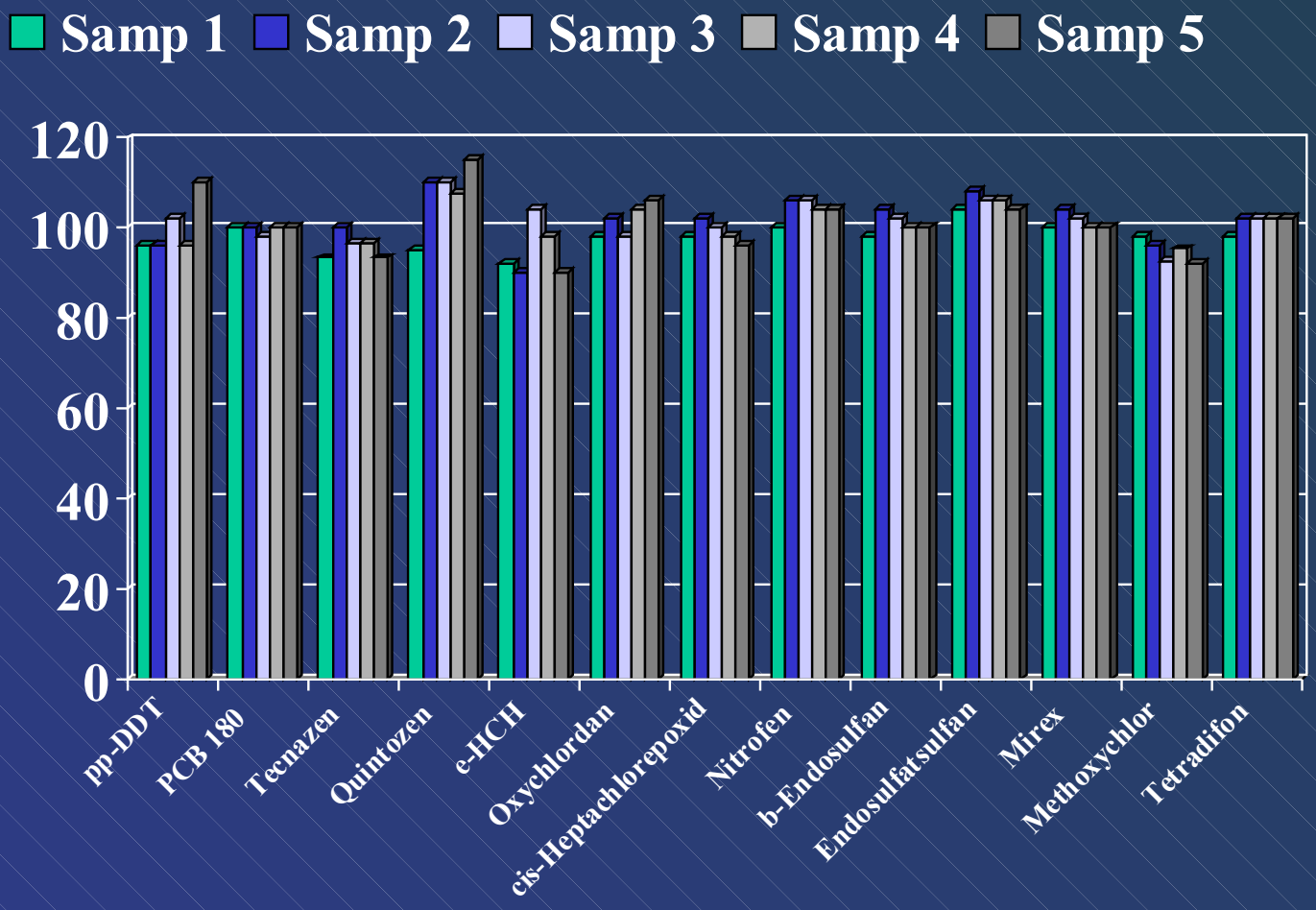
S19 Replicates OCP -1



S19 Replicates OCP - 2



S19 Replicates OCP - 3



Conclusions

Acknowledgments

- Michael Fluornoy and George Burkett, Severn Trent Labs, Sacramento, CA
- John Murray, GuaranTek, Denver, CO
- Dr. Armin Trenkle, Landwirtschaftliches Technologiezentrum Augustenberg, Karlsruhe, Germany