KANTONALES LABOR ZÜRICH

Mineral oils depicted by gas chromatograms

Principals for the analysis in food

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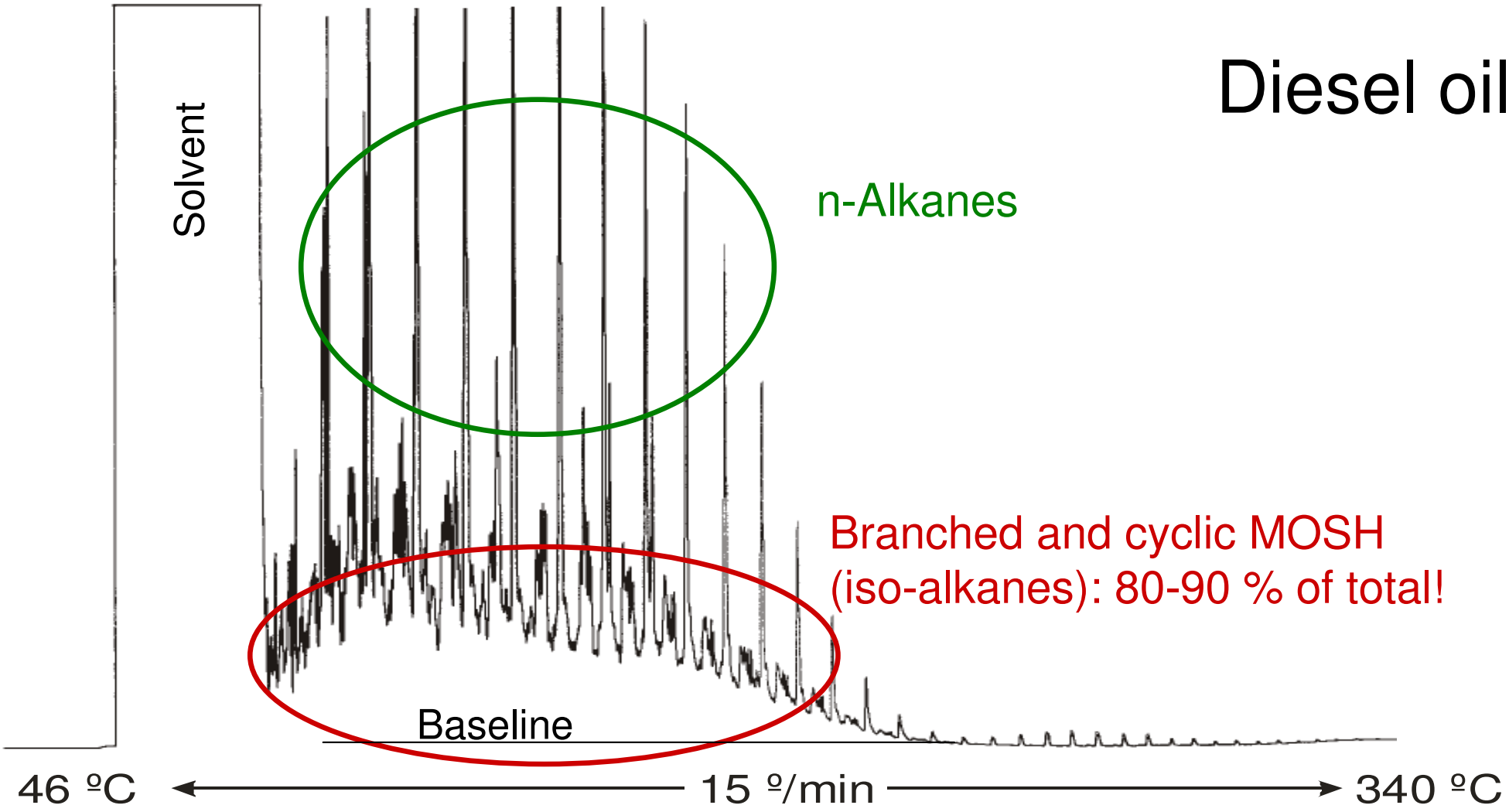
Mineral oils in food packaging – developments and solutions
Seminar Berlin 23rd of September 2011

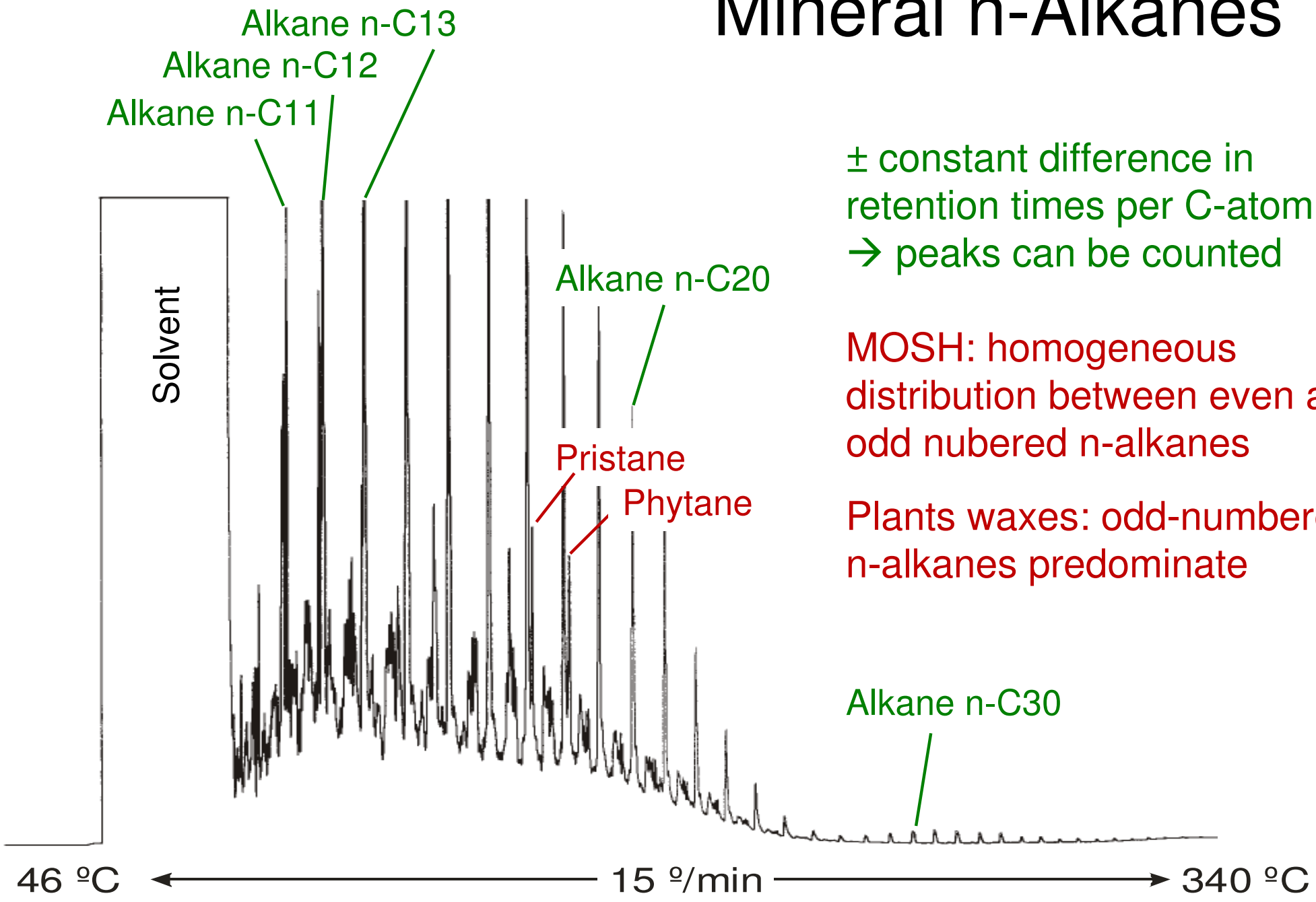
GC of mineral oil saturated hydrocarbons (MOSH)

GC on apolar stationary phase separates ~ by volatility, ~ by molecular mass



Gas chromatography of mineral oils

Mineral n-Alkanes



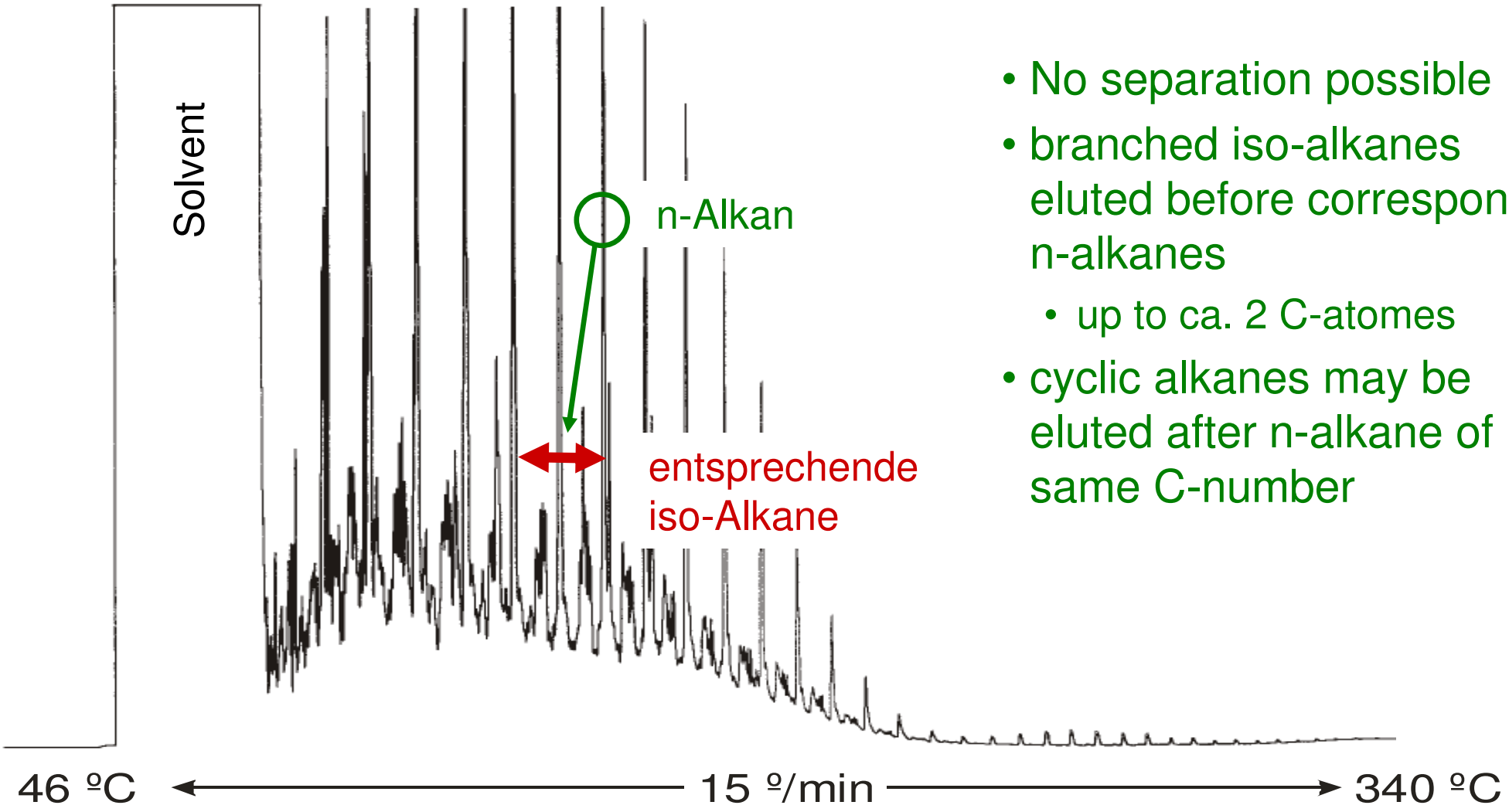
± constant difference in retention times per C-atom
→ peaks can be counted

MOSH: homogeneous distribution between even and odd nubered n-alkanes

Plants waxes: odd-numbered n-alkanes predominate

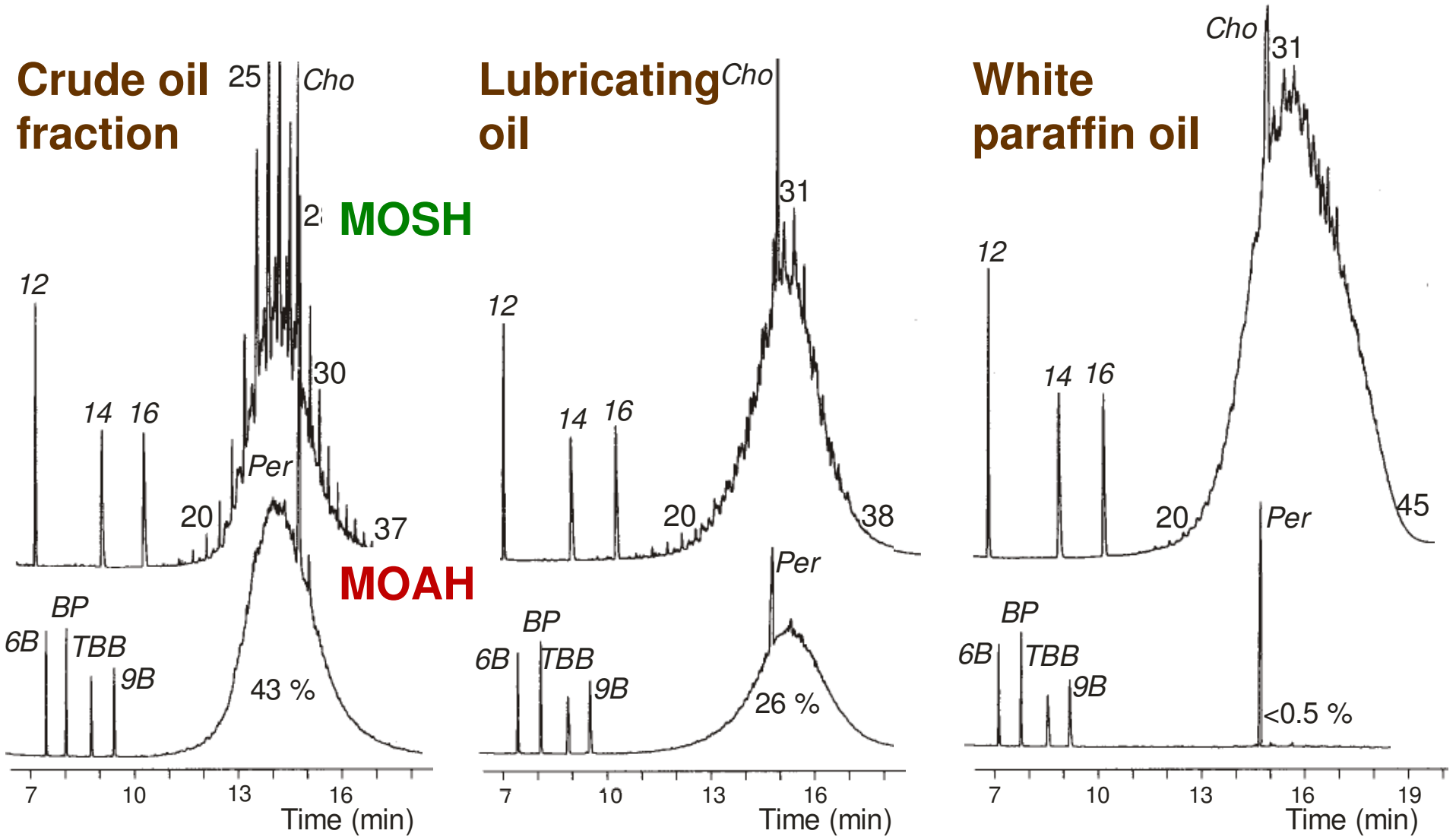
Mineral iso-alkanes

(separation on a 10 m x 0.25 mm i.d. column)



- No separation possible
- branched iso-alkanes eluted before corresponding n-alkanes
  - up to ca. 2 C-atomes
- cyclic alkanes may be eluted after n-alkane of same C-number

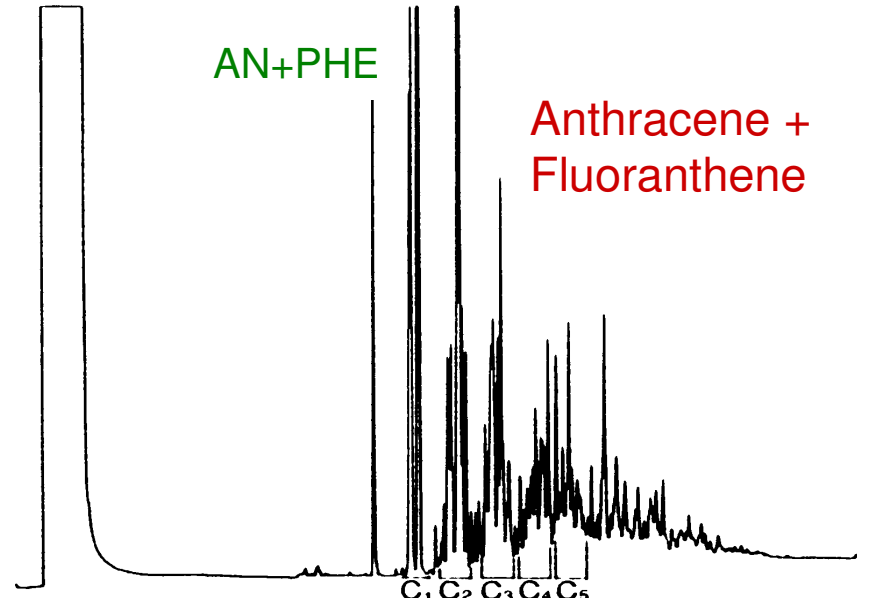
Mineral oil aromatic hydrocarbons (MOAH)

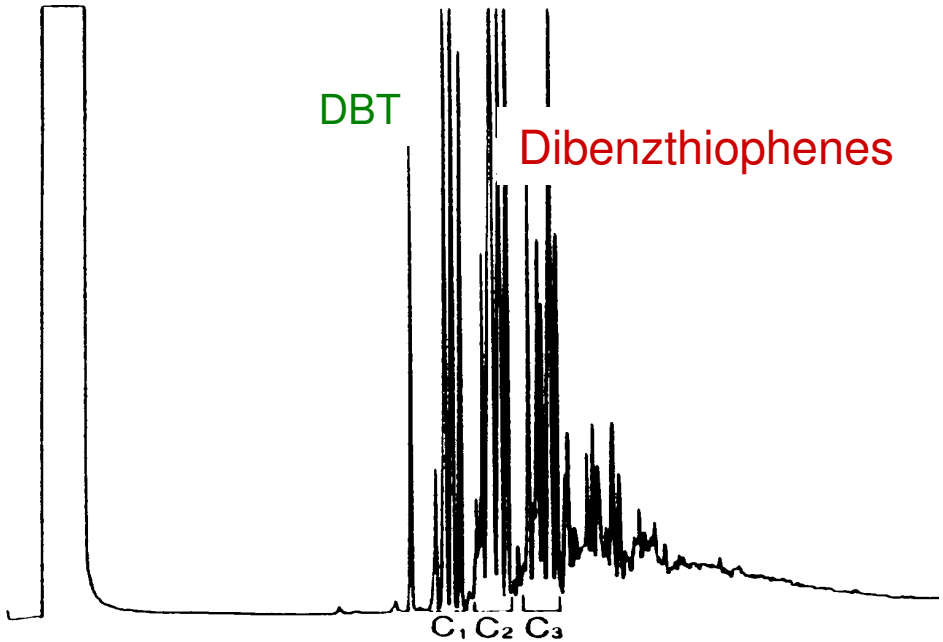


On non-polar stationary phases MOAH are coeluted with MOSH
but carbon number does not correspond:
Methyl anthracene (C_{15}) eluted at n-C_{21}, Chrysene (C_{18}) at n-C_{27}, Pyrene (C_{16}) at n-C_{24}

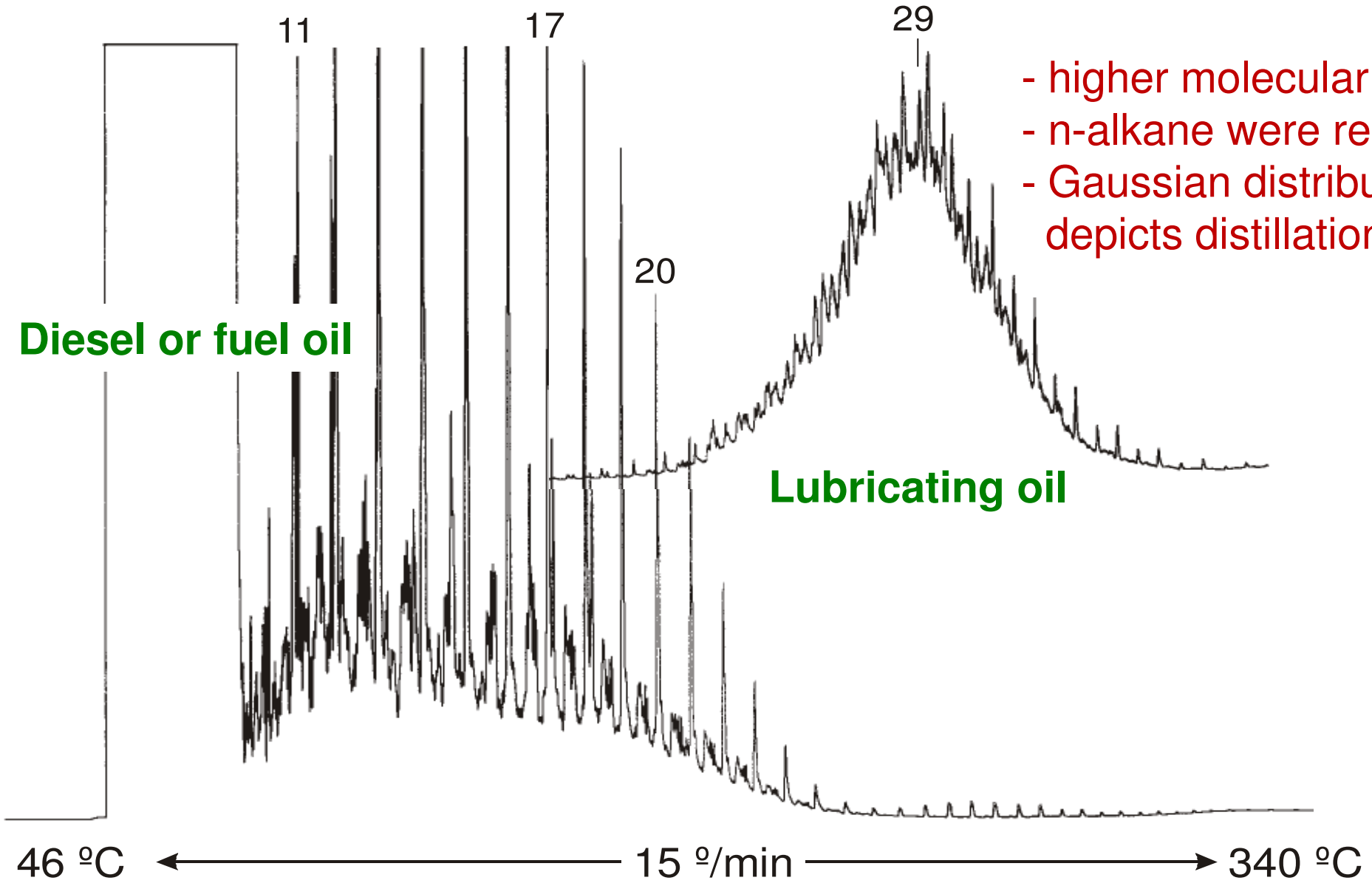
Comparison of MOAH with PAHs

- Polycyclic aromatic hydrocarbons (PAHs)
  - from pyrolysis processes (coaking, smoked products, black saussages…)
  - little alkylated compounds
  - widely analyzed for decades
  - analyzed (and evaluated) as individual substances
- MOAH
  - from geological processes
  - 97 to >99 % alkylated
  - very many isomers
  - cannot be resolved to individual substances → hump
  - little investigated (and evaluated)



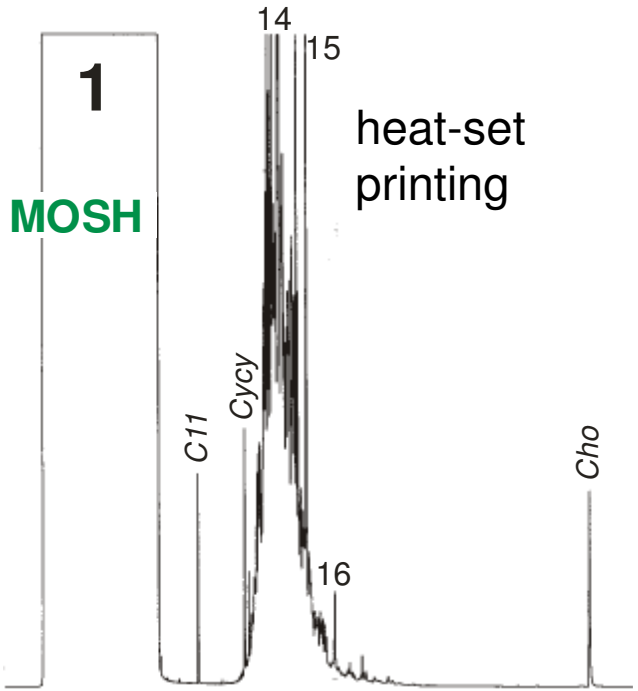


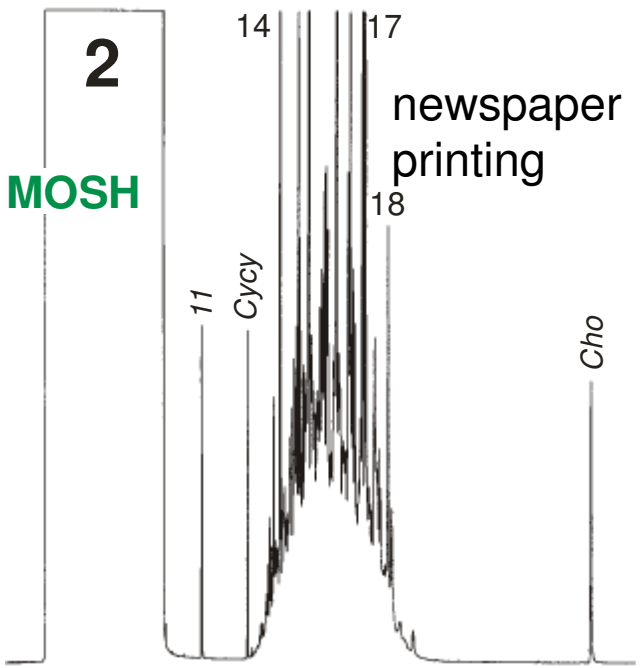
Characterization of mineral oil products

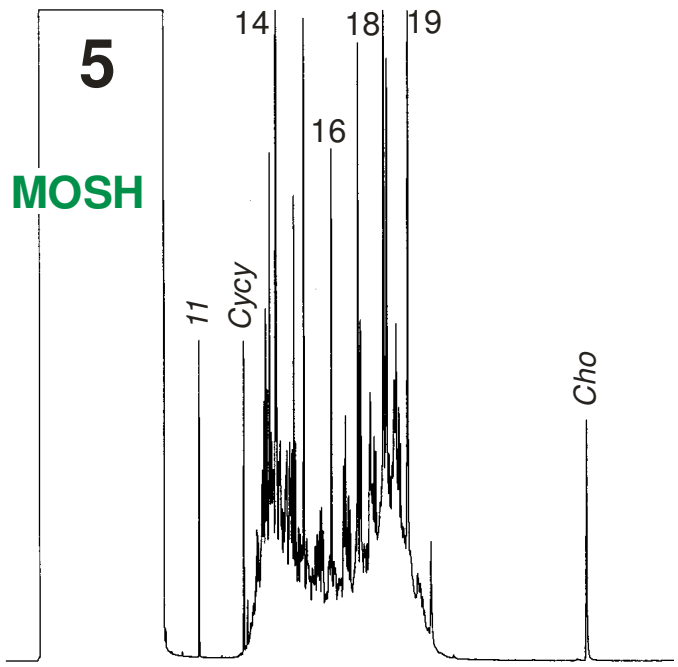


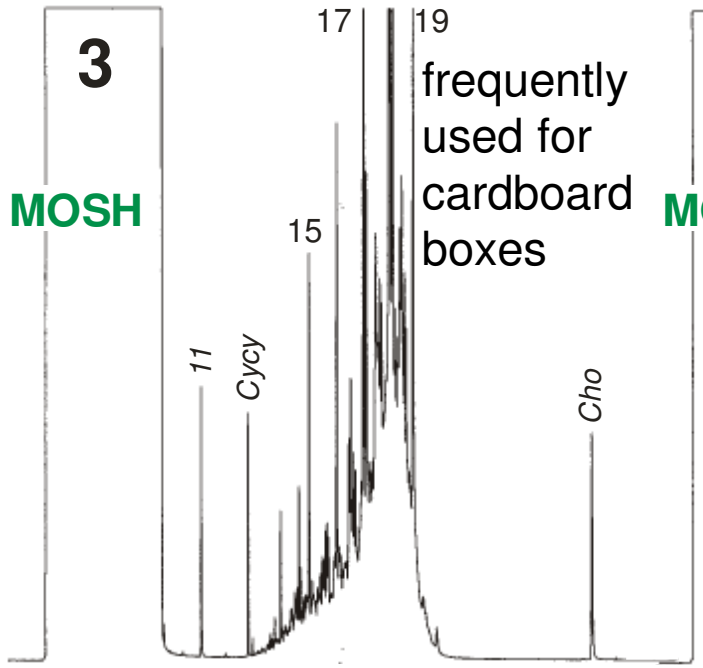
Pictures of mineral oil products

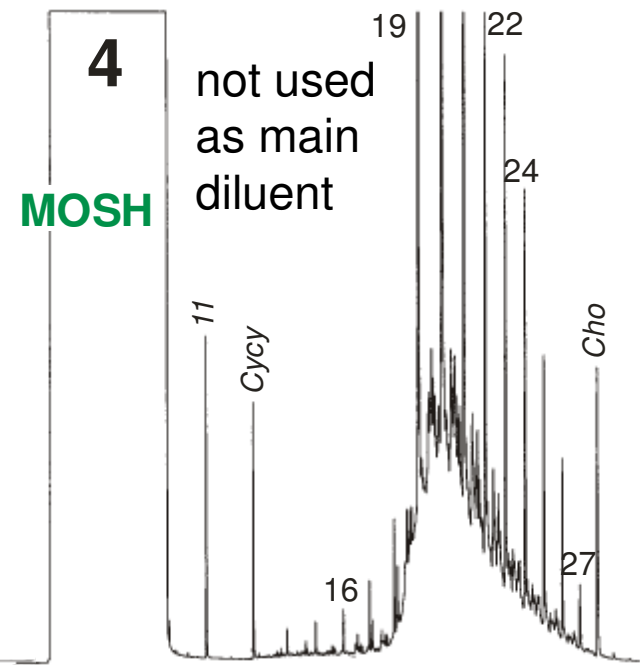
Oils for printing inks

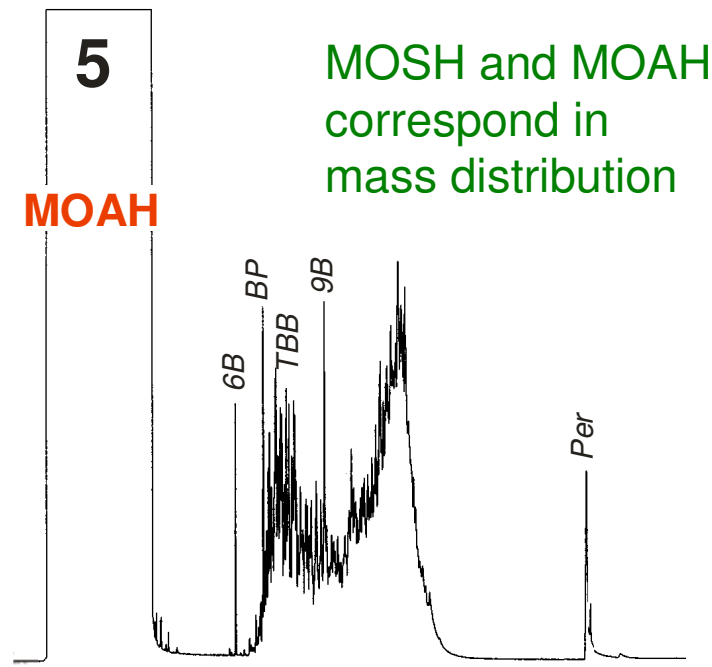












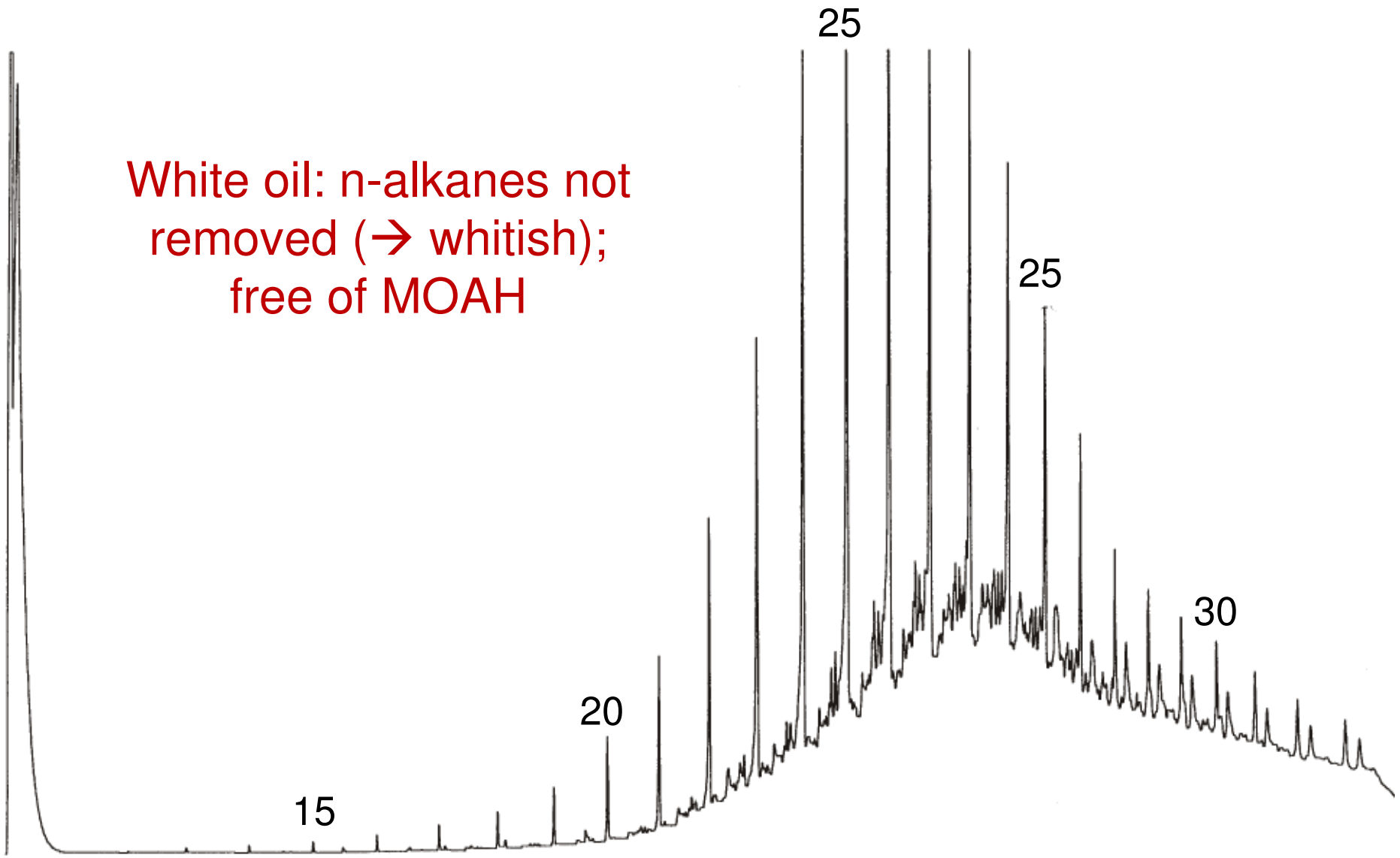
Batching oil for jute bags

Contaminated hazel nuts, cocoa beans, coffee, rice, oil seeds

Vaseline

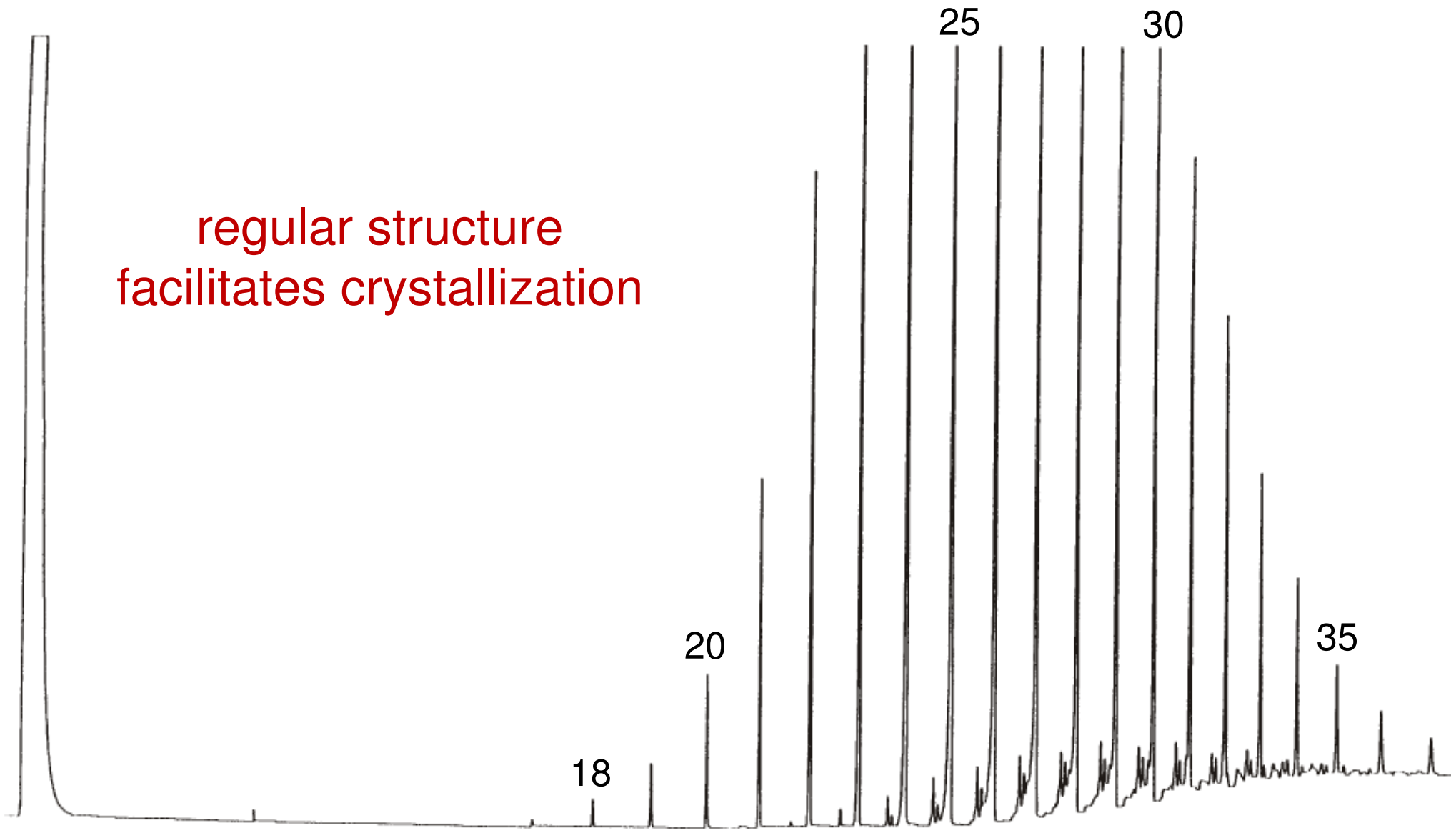
sold, e.g., for cosmetics or pharmaceutical applications

White oil: n-alkanes not removed (→ whitish); free of MOAH

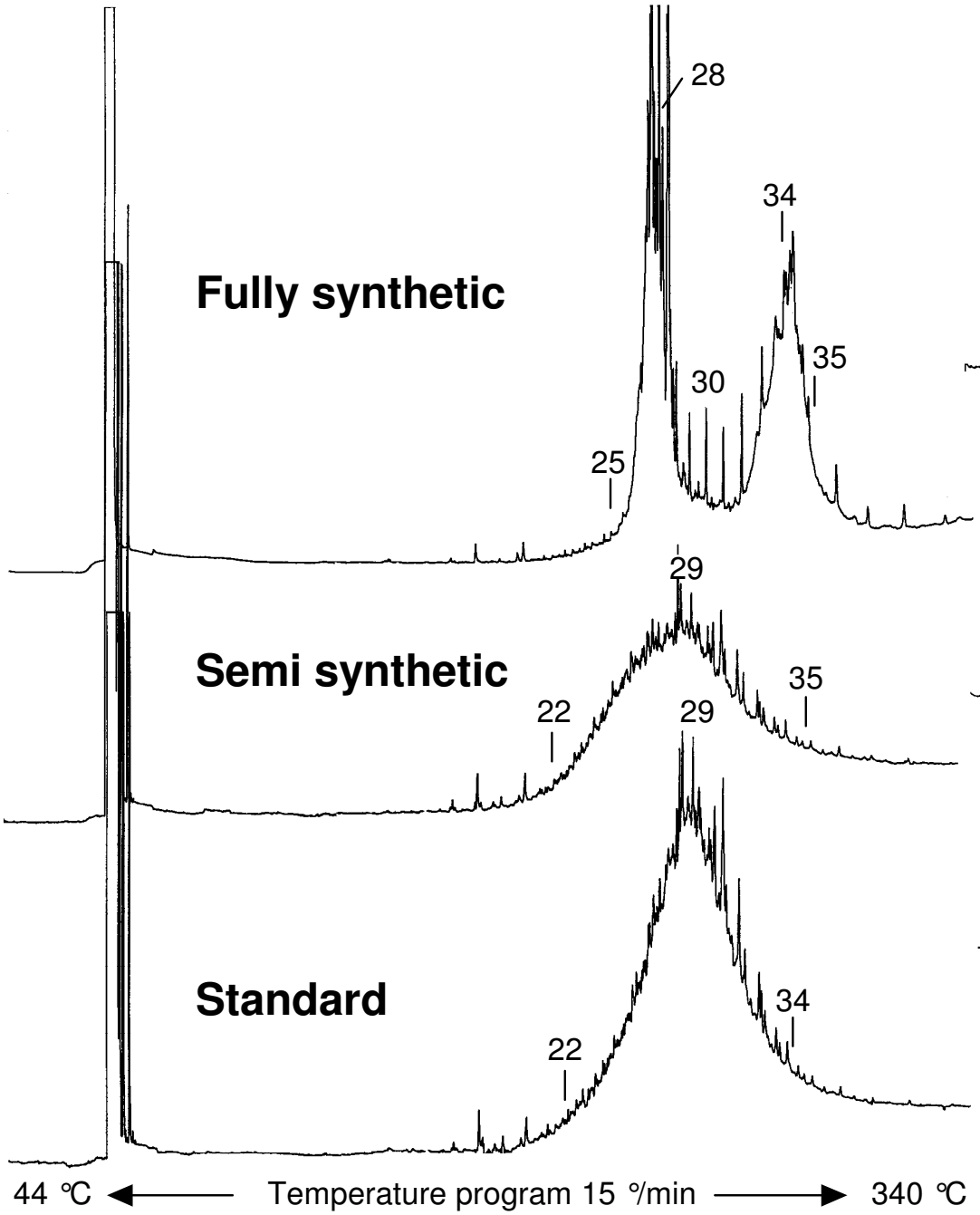


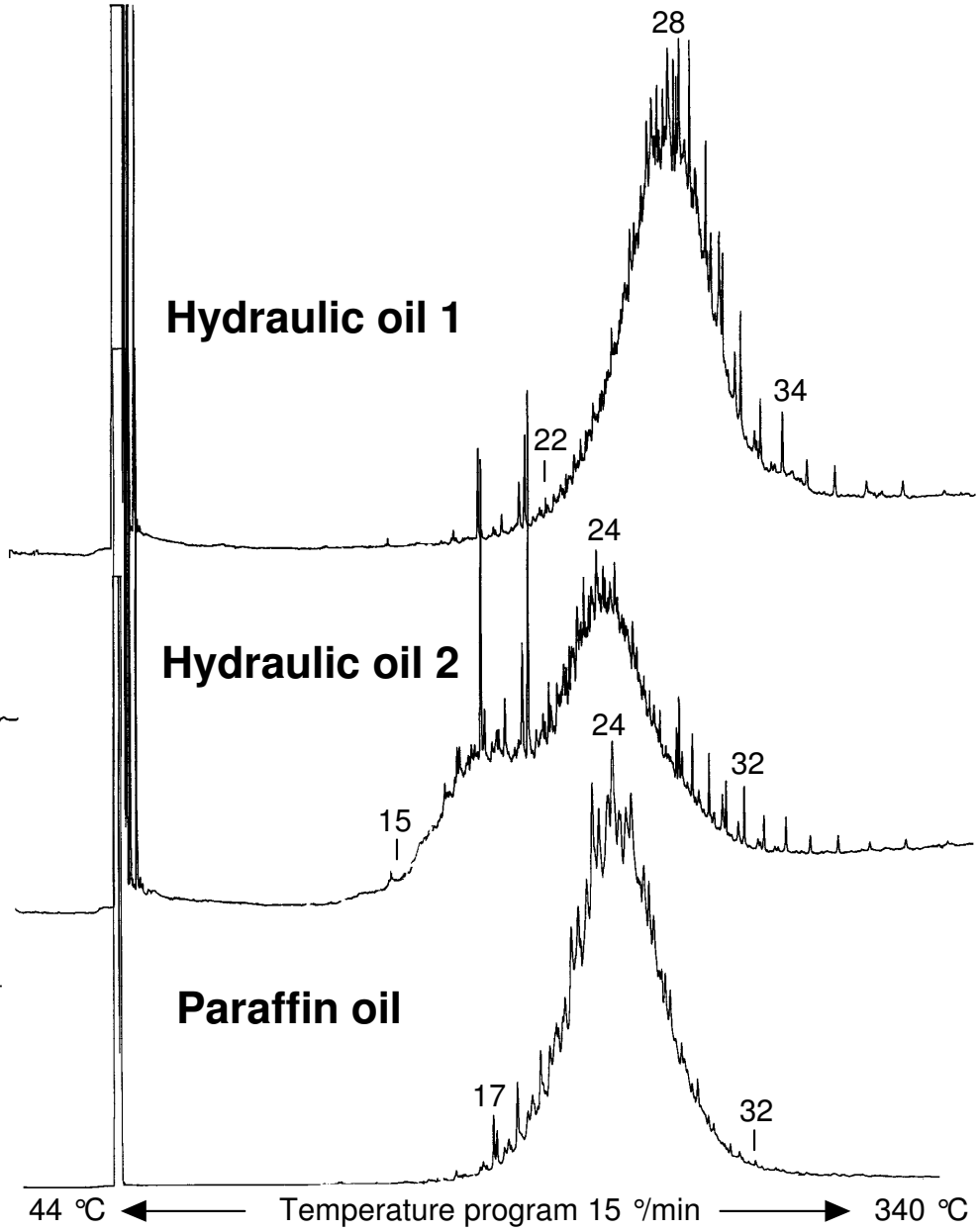
Wax (candle)

regular structure
facilitates crystallization



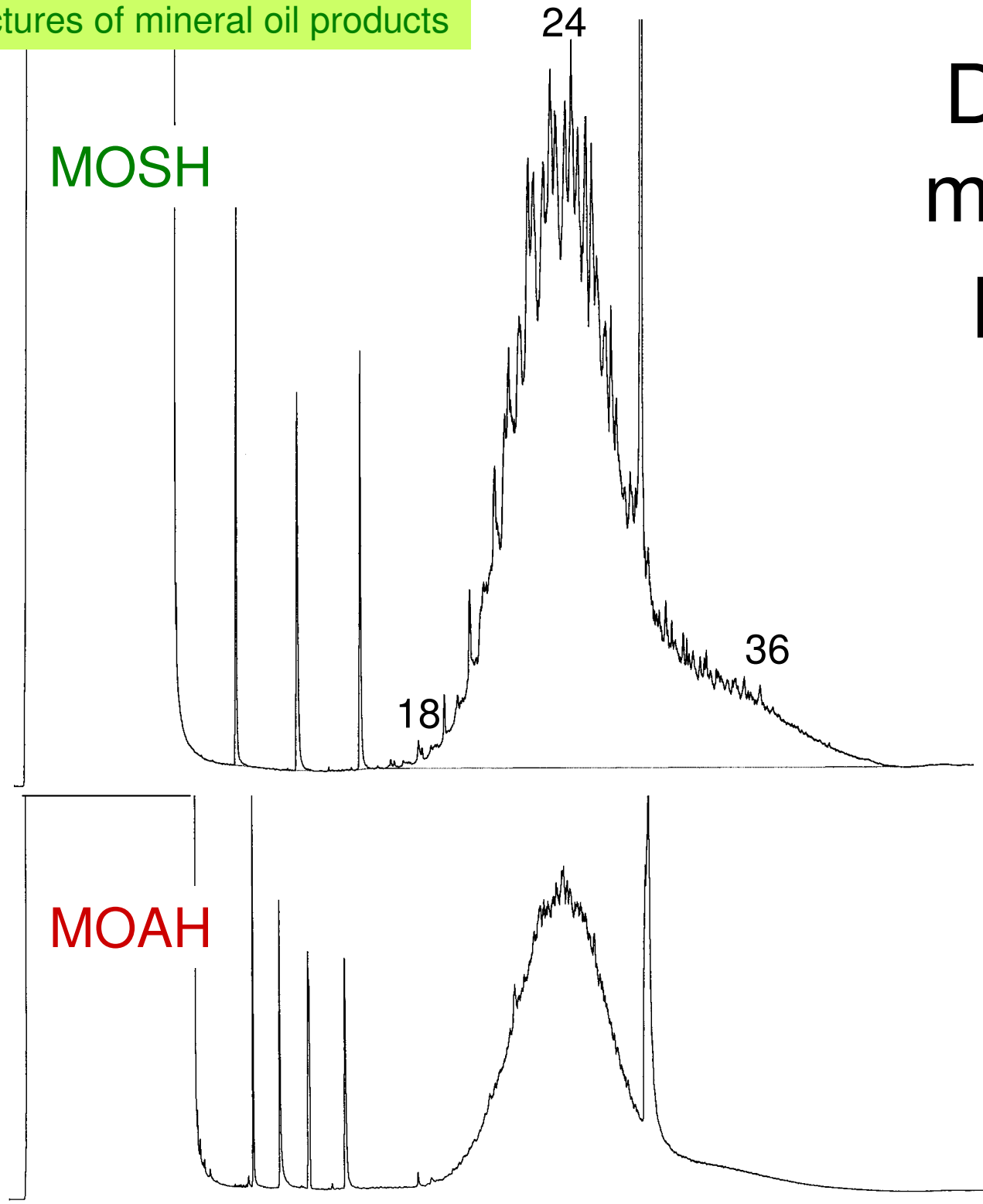
Lubricating oils





Hydraulic oils

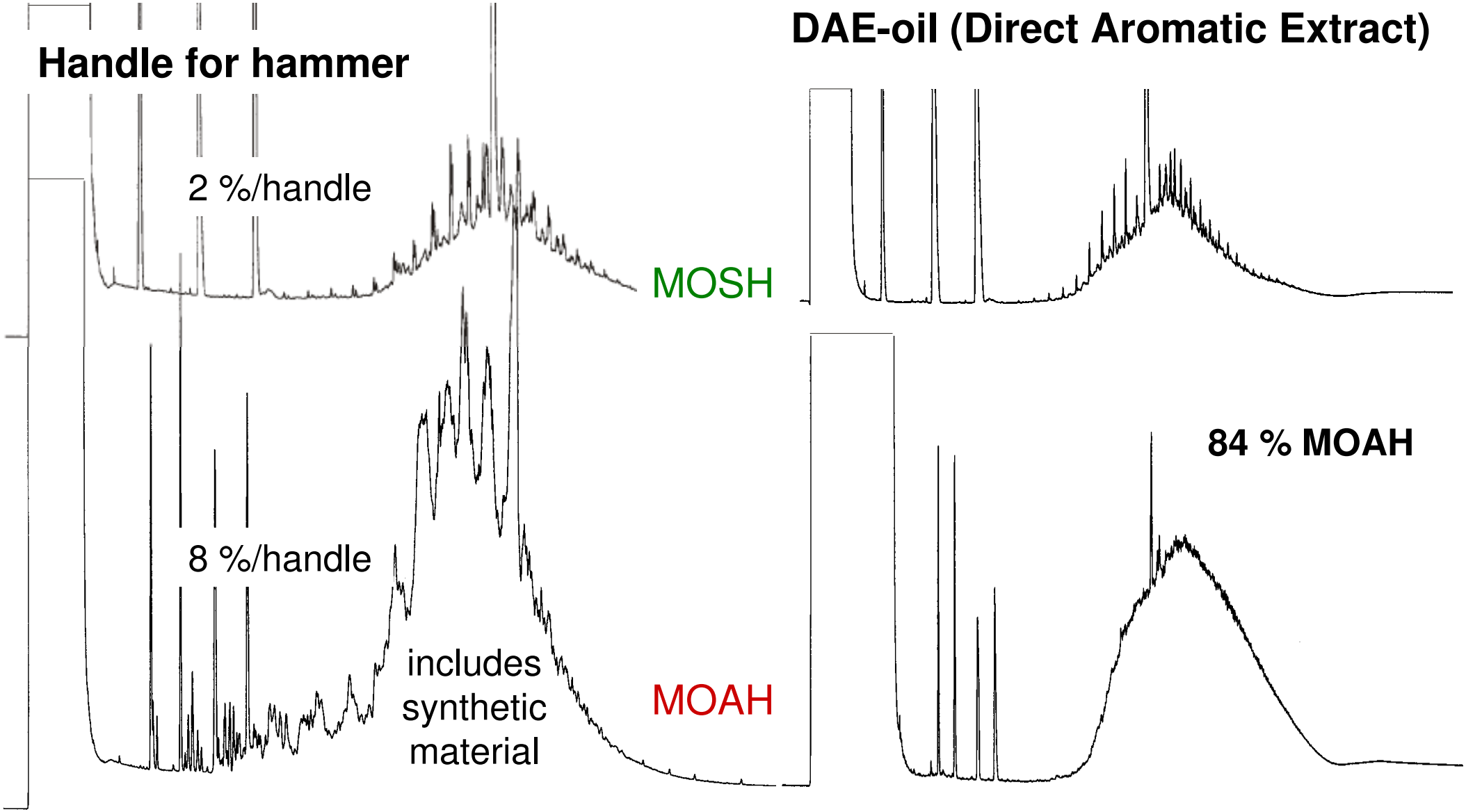
Pictures of mineral oil products



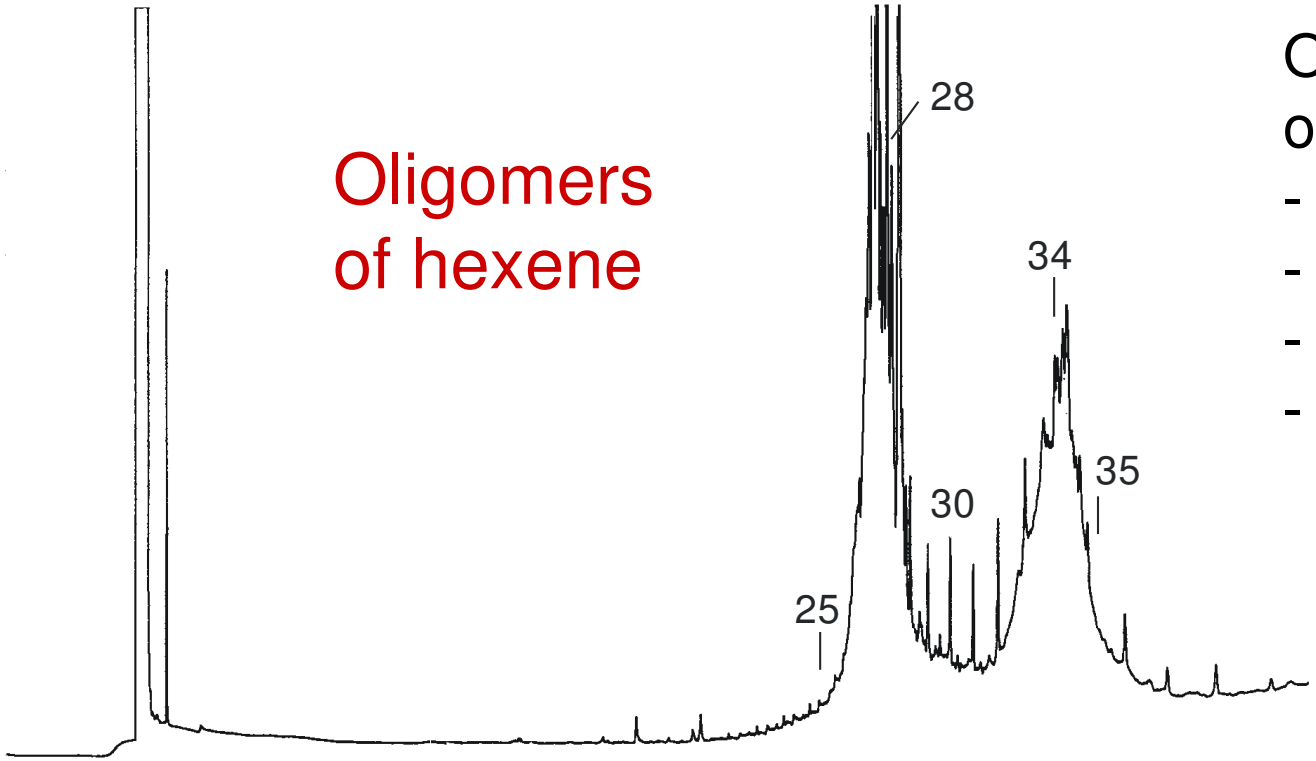
Defoamer for manufacturing paperboard

mostly replaced by fatty acid ethoxylates

Extender oils (plasticizing rubber and elastomers)



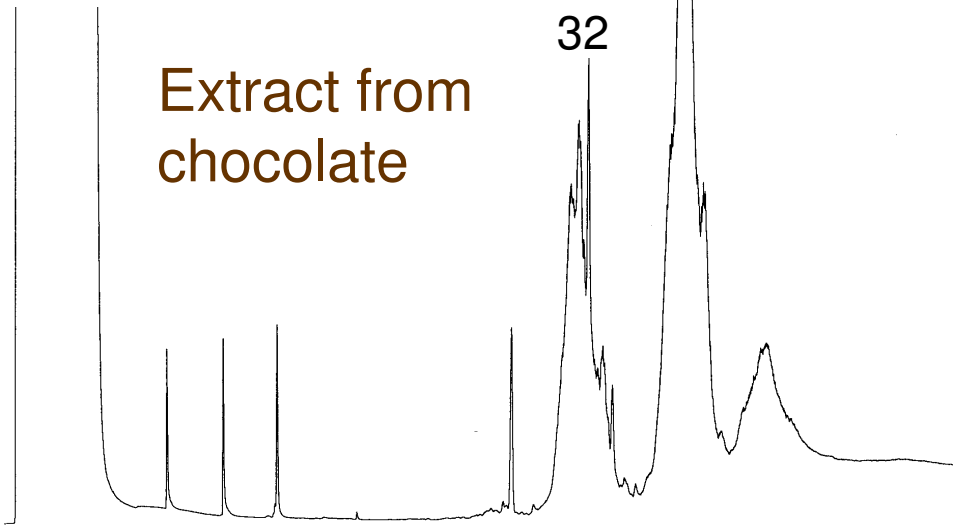
Poly-alpha-Olefines (PAOs)



Oligomers
of hexene

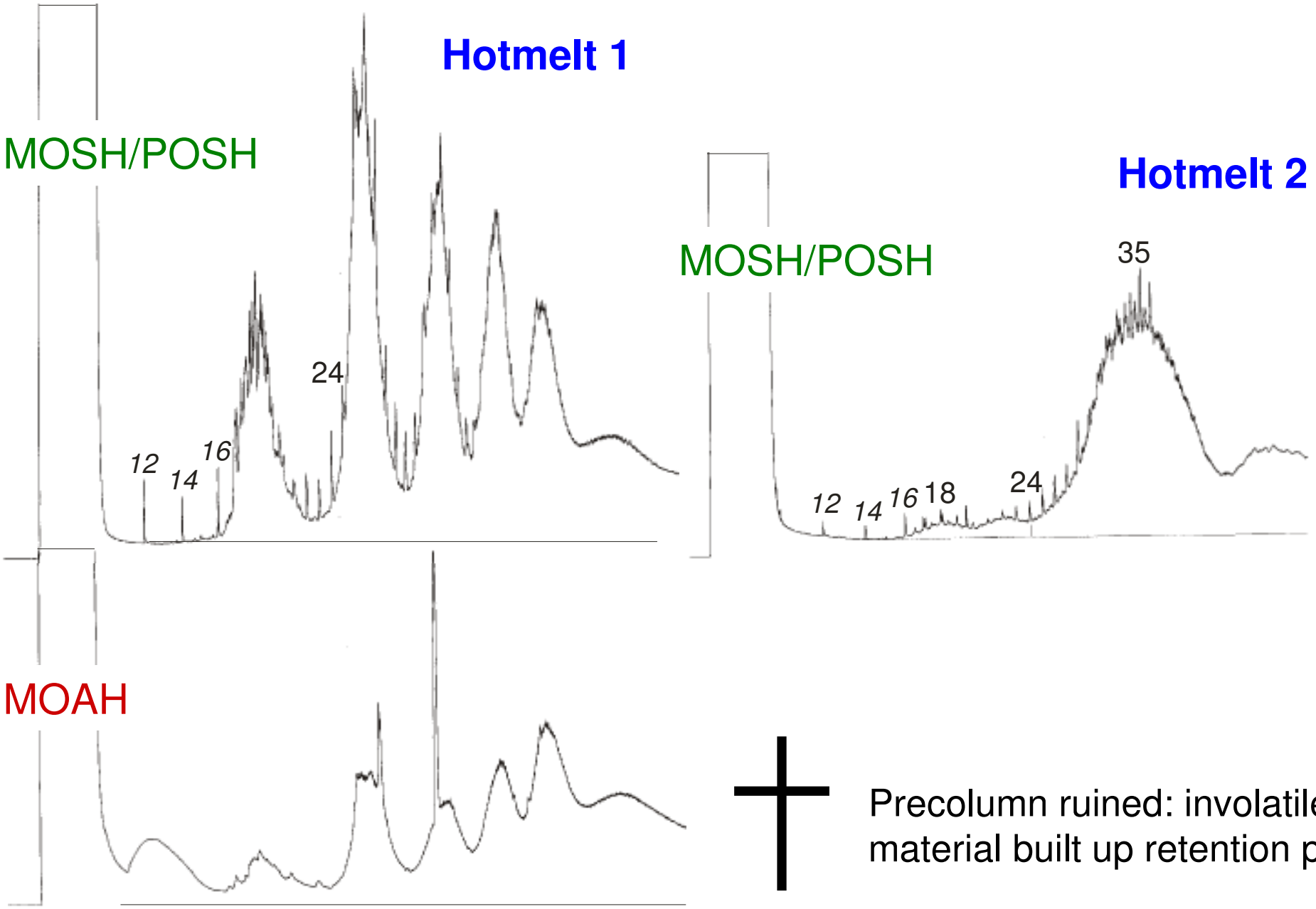
Oligomers of synthetic olefins
or cracking products
- synthetic lubricating oils
- „food-grade“ oils
- adhesives
- cosmetics

PAO are free of MOAH



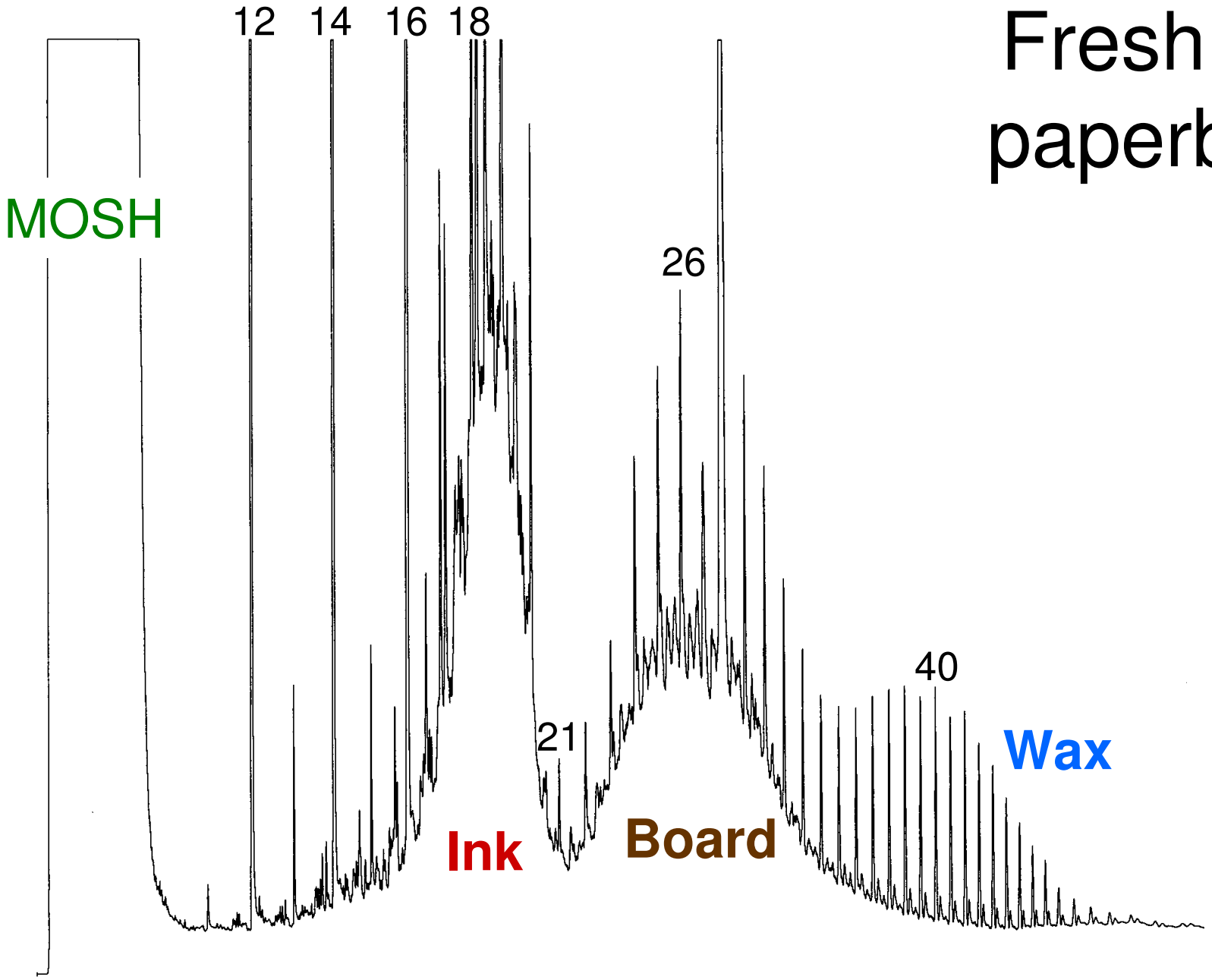
Extract from
chocolate

Adhesives (hotmelts) for paperboard



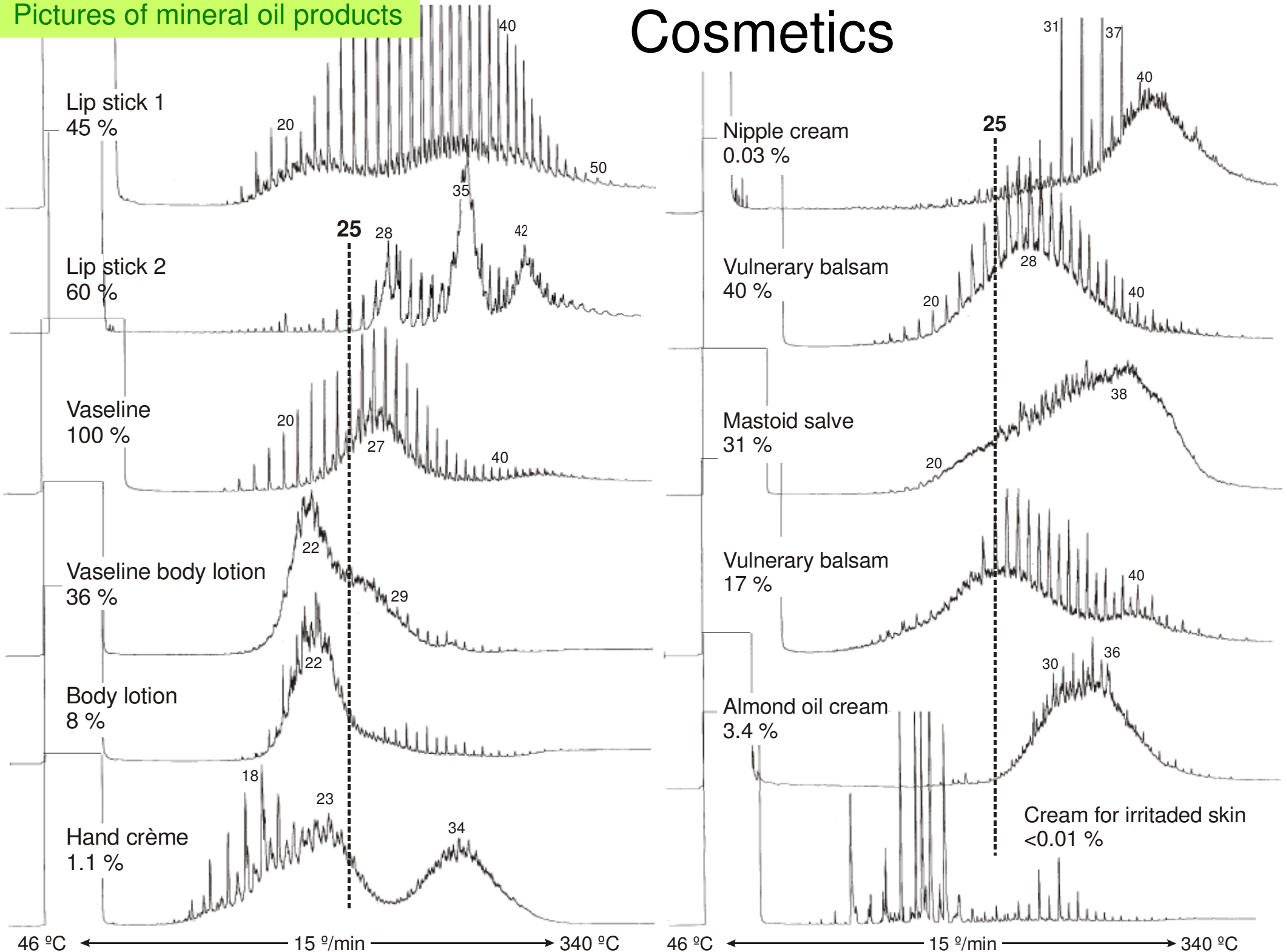
† Precolumn ruined: involatile material built up retention power

Fresh fiber paperboard



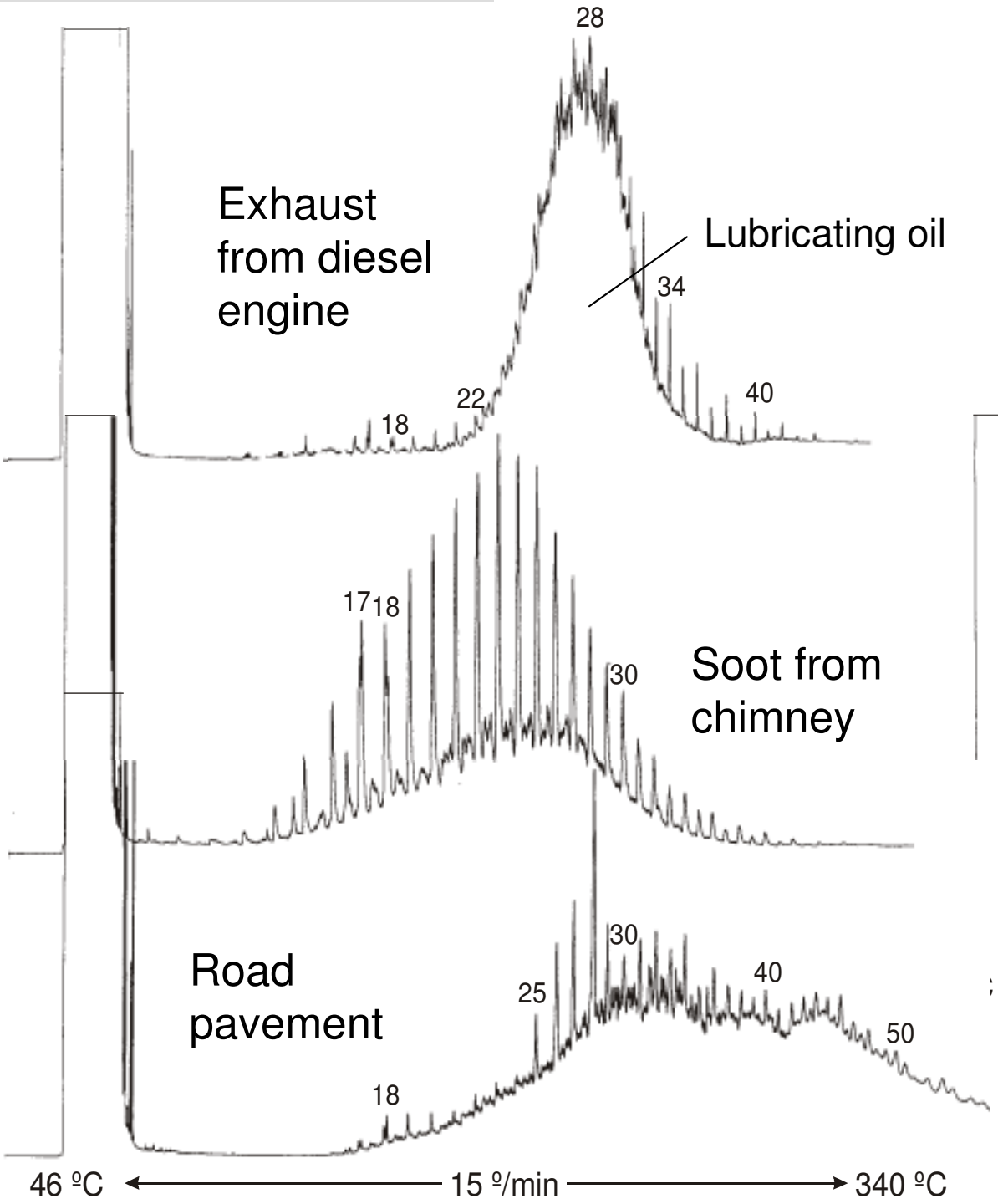
Pictures of mineral oil products

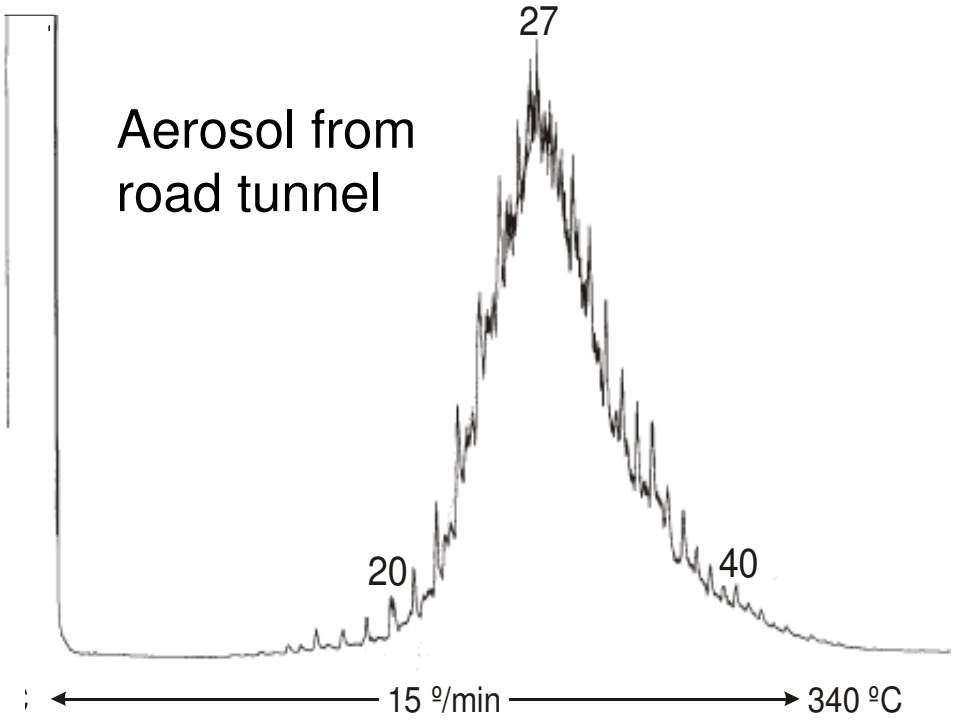
Cosmetics



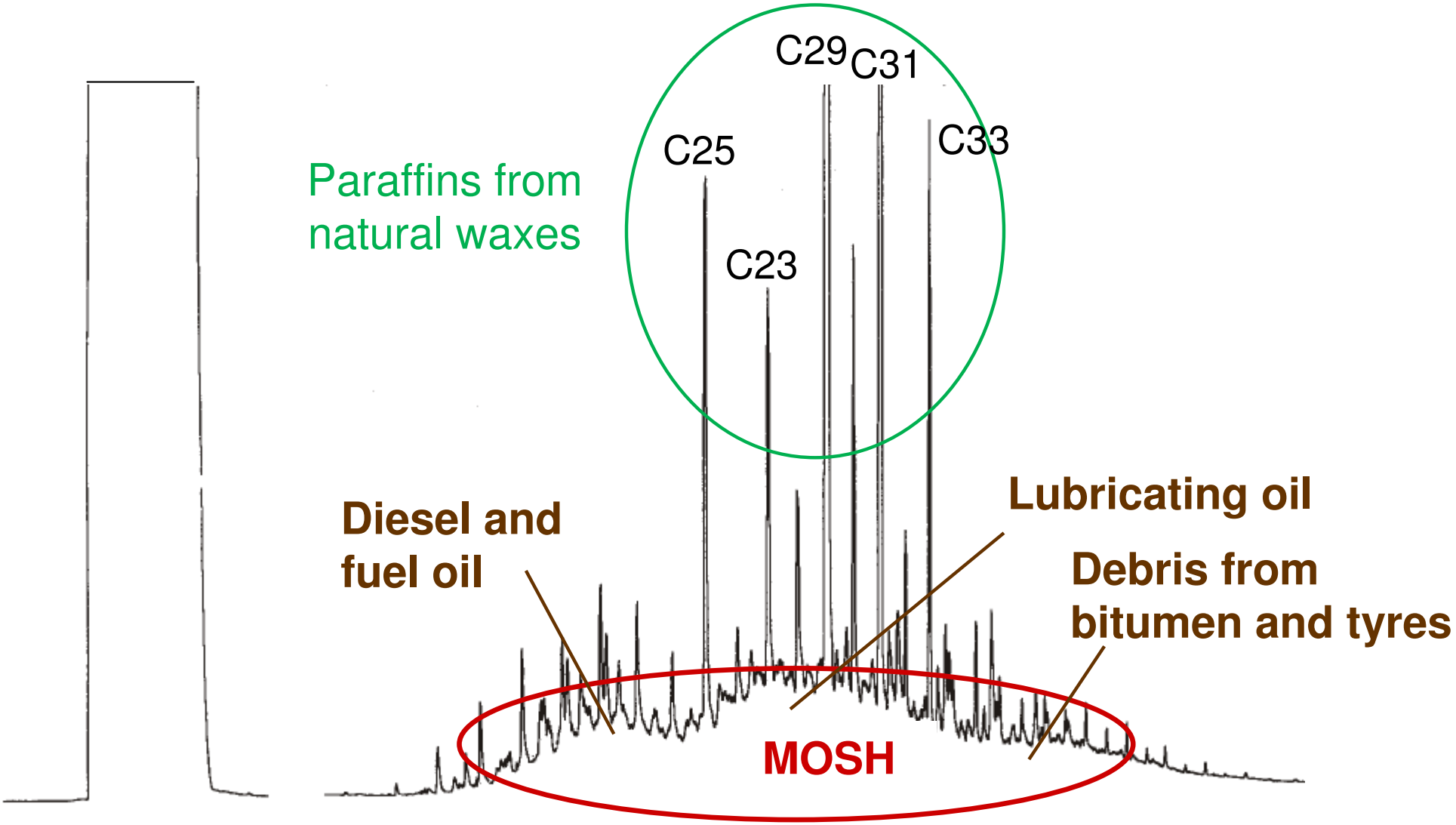
Mineral oils in background

Environmental contributions

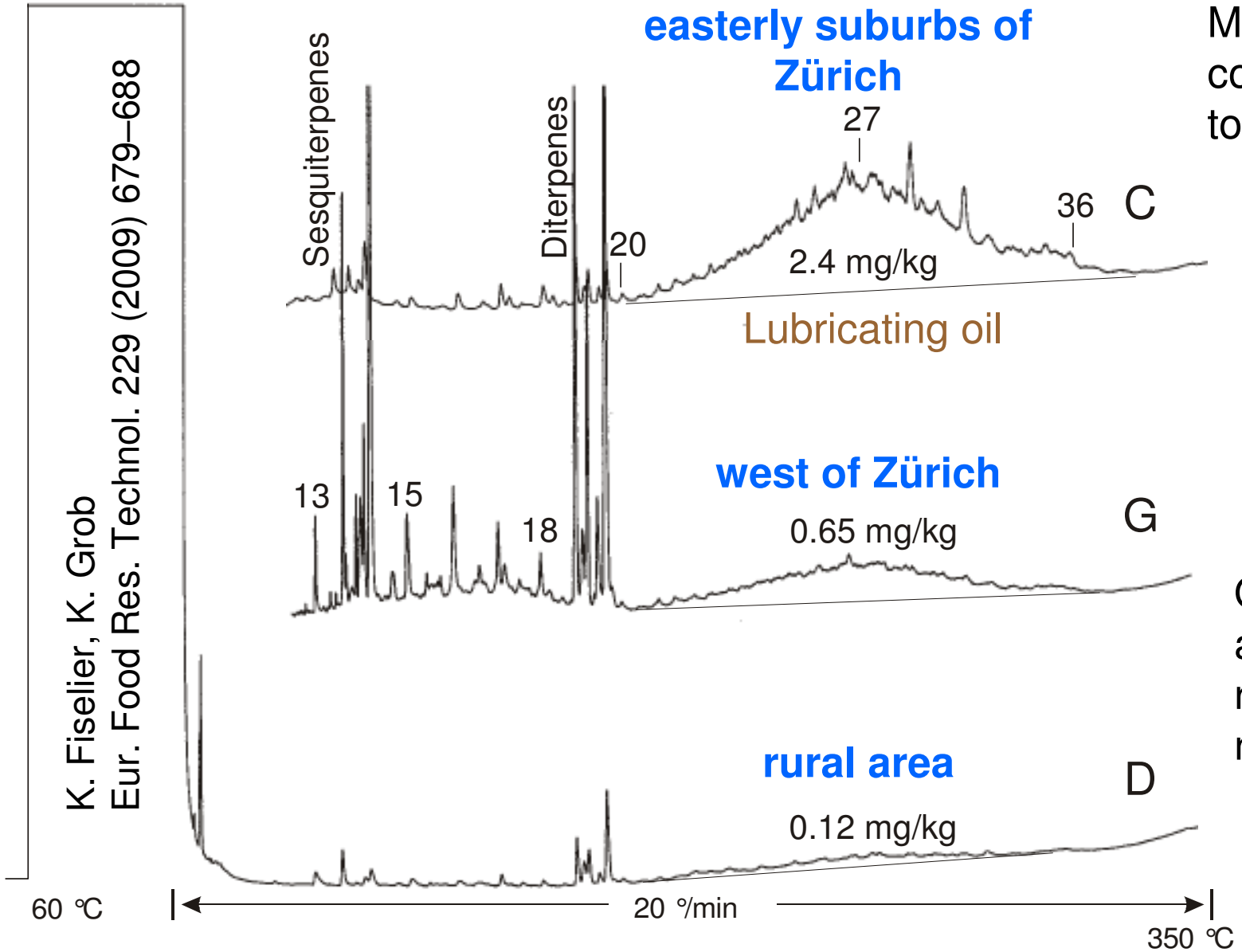




MOSH in wheat



MOSH in sunflower seeds: environmental contamination

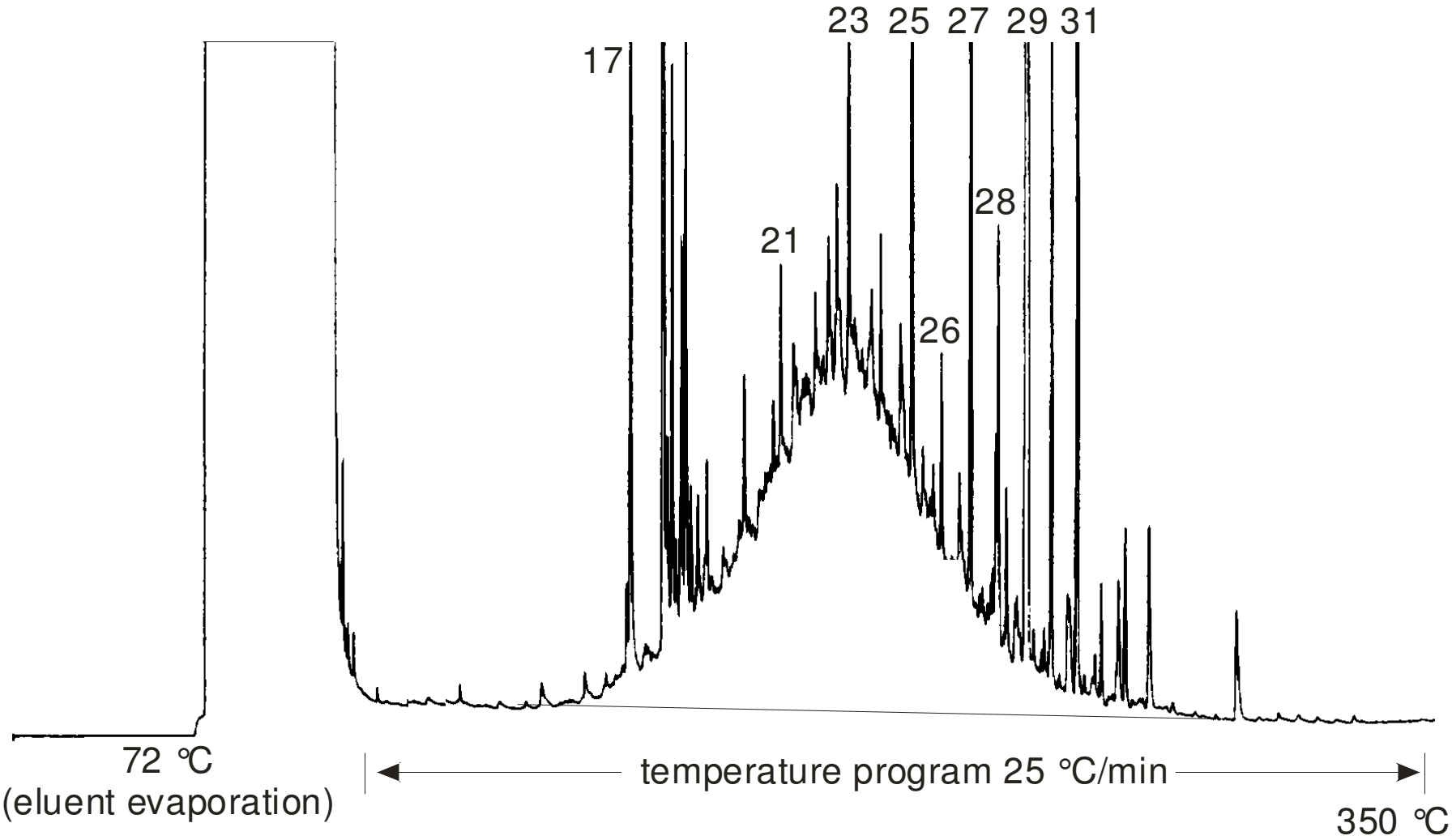


Manually picked seeds; concentrations referring to oil

On-line HPLC-GC-FID after enrichment and removal of long-chain n-alkanes

MOSH in human body fat

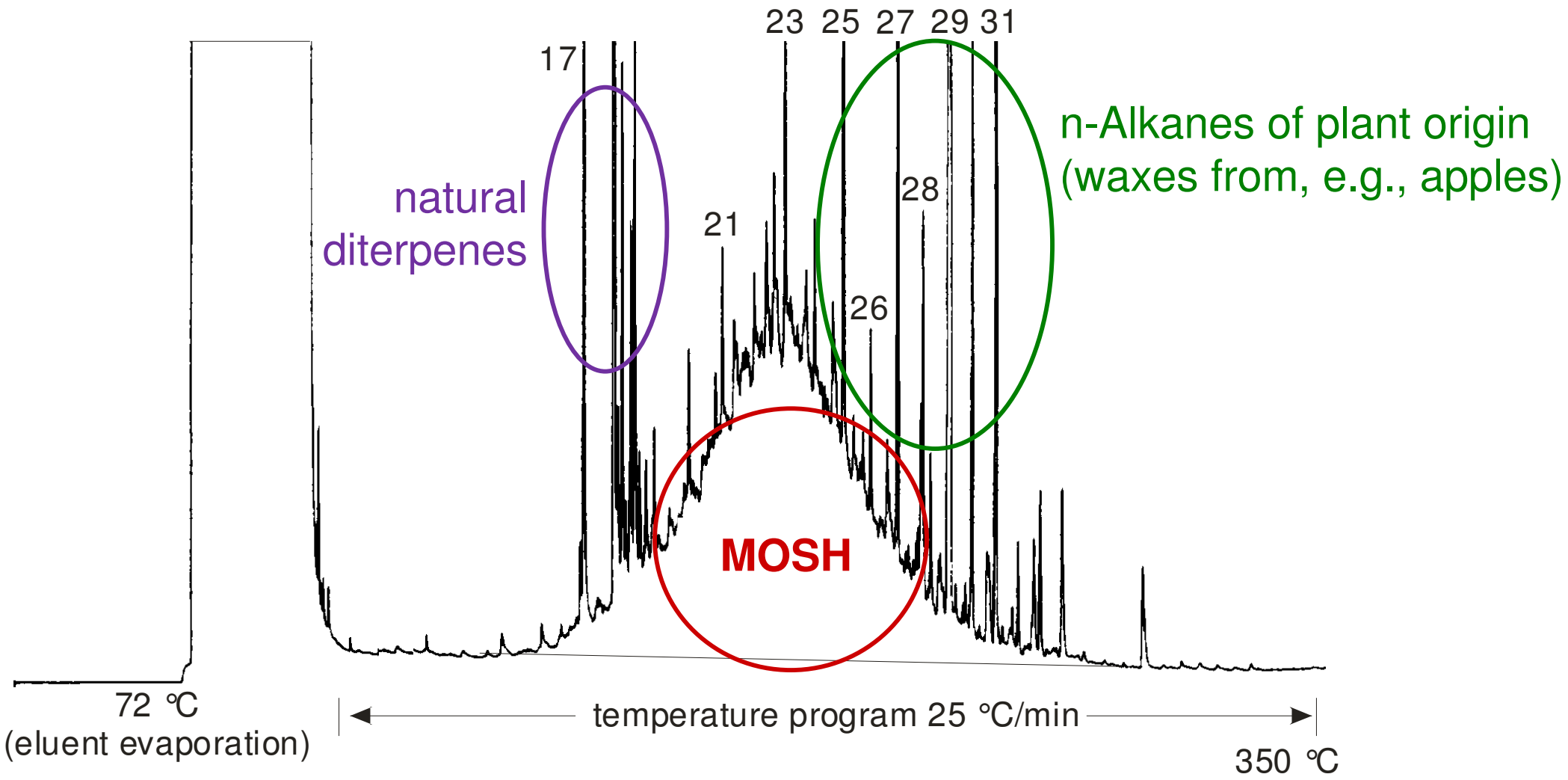
Abdominal fat obtained by Caesarean sections



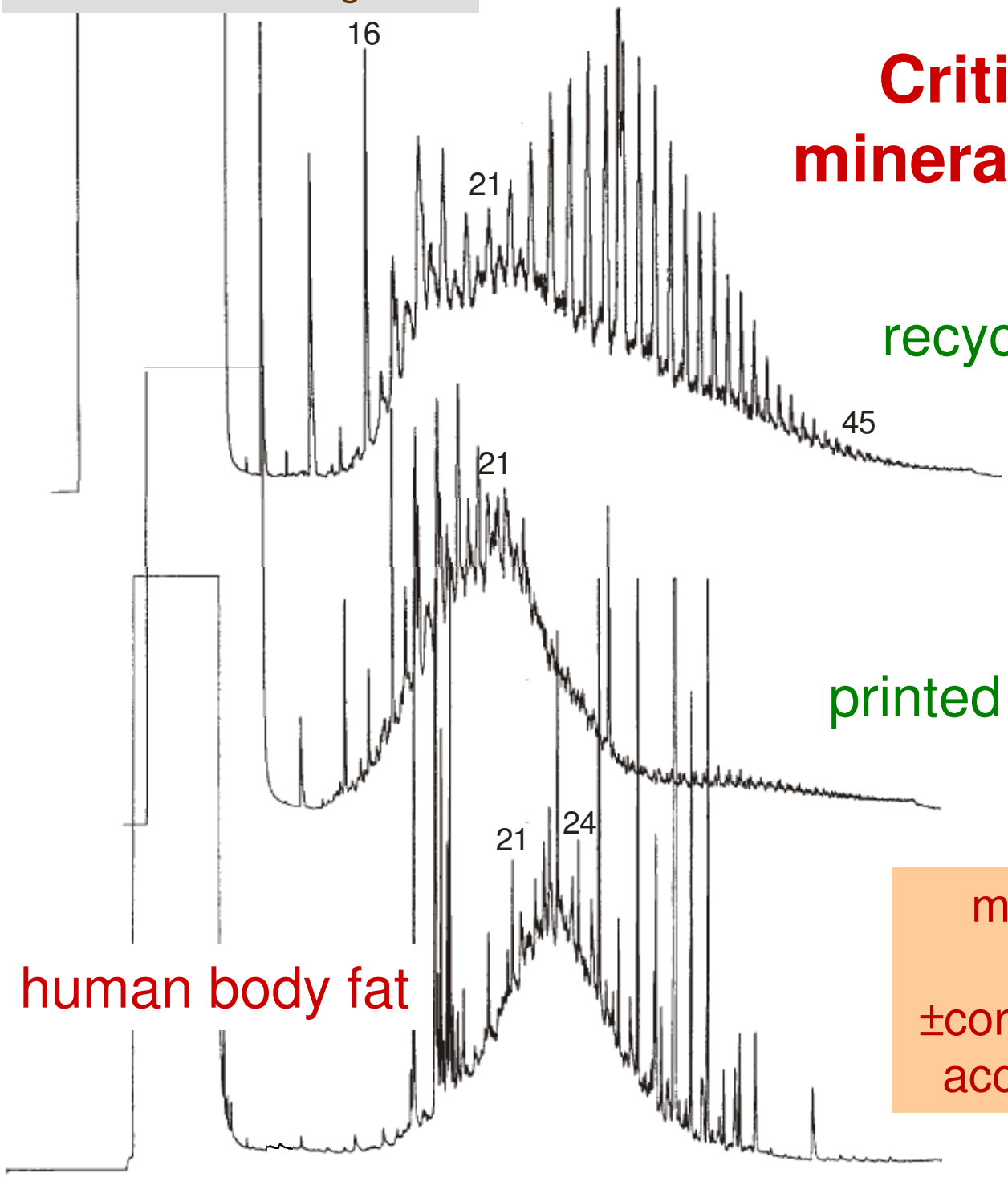
Mineral oil paraffins in human body fat and milk. N. Concin, G. Hofstetter, B. Plattner, C. Tomovski, K. Fiselier, K. Gerritzen, S. Fessler, G. Windbichler, A. Zeimet, H. Ulmer, H. Siegl, K. Rieger, H. Concin, K. Grob. Food and Chemical Toxicology, 46 (2008) 544-552

MOSH in human body fat

tells us about the critical hydrocarbons: those our body is unable to eliminate



Mineral oils in background



Critical composition of mineral oils for printing inks

recycled board

printed newspaper

human body fat

molecular mass distribution of MOSH in printing inks ±corresponds to that absorbed and accumulated by the human body

Principals for the analysis

- Tasks of analysis
- Extraction from packaging material and foods
- Aliquot required for analysis in foods
- FID as detector
- Information obtained from GC analysis

Tasks of the analysis

- Mineral oil as MOSH and MOAH, separated from endogenous hydrocarbons
- **MOSH** fractions in packaging (paperboard and plastic):
  - <C16
  - C16-C24 (limit for migration through gas phase)
  - C24-C35 (only for applications with wetting contact)
- **MOSH** fractions in food:
  - <C16 (envisioned limit: 12 mg/kg)
  - C16 to about C25 (migration through gas phase)
  - C16-C35 (envisioned limit: 0.6 mg/kg)
- **MOAH** fractions in packaging:
  - <C24 (no separation at C16)
  - C24-C35 (only for applications with wetting contact)
- **MOAH** in food:
  - up to about C25 (migration through gas phase)
  - up to C35 (envisioned limit: 0.15 mg/kg)

POSH: same as MOSH until evaluated otherwise?

Critical points in sampling and storage of samples

Information needed: history of the sample? conditions of storage

- migration from transport box?
- reference samples?

Migration is a continuing process

- measurement for moment of sampling? → separate different parts
  - box and bag into aluminum foil
  - food in glass jar, lid protected by aluminum foil
- further storage? → wrap in aluminum foil (no evaporation outwards, no contamination from outside)

Paperboard: sample not from top of stack

- pick-up from air, loss into air

Sending sample

- paperboard: wrap into aluminum foil

Extraction of dry foods

- Problem: inclusions into solid structures!
- Experiments on dry noodles (no eggs)
  - Migrate from paperboard completely extracted overnight/RT or 2 h/60 °C (<10 % in 2nd extract weekend/60 °C)
  - MOSH worked into noodles during manufacture little extracted
    - tested for noodle prepared in the lab
  - complete extraction requires swelling with water (cooking)
- Powdered baby formula, milk powder
  - overnight/RT: ca. 30 % of migrate, but <10 % of fat and endogenous hydrocarbons
  - overnight/60 °C: complete for migrate, 25 % for fat, endogenous hydrocarbons and MOSH in the sample before packing
    - preferred conditions: reduces interference
  - complete extraction: heating in concentrated HCl (30 min/80 °C)

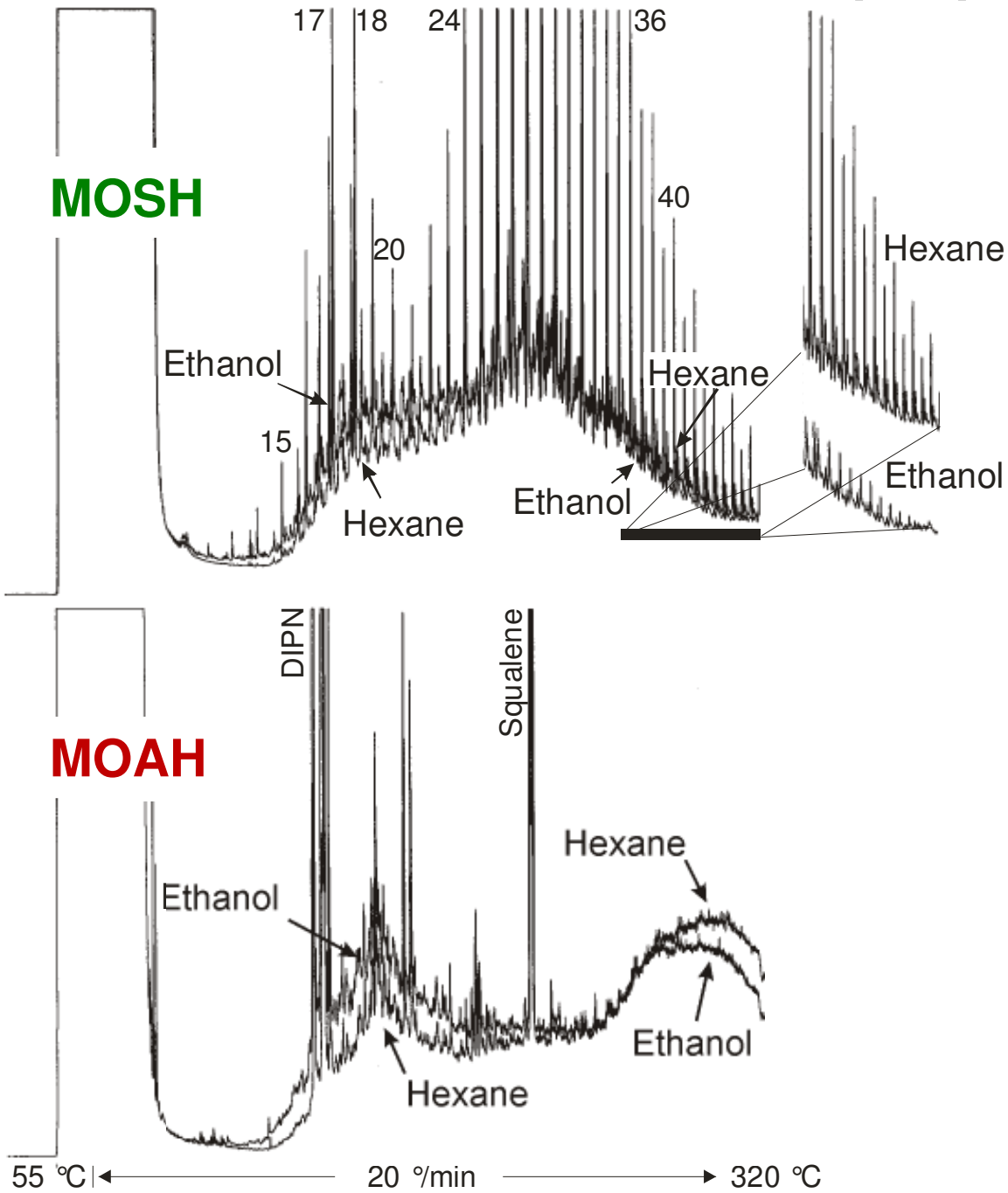
Extraction yields for solids

- cannot be checked by spiking samples!
  - added material is easier to extract than that included
- Second extracts
  - rinse solid residue after first extraction (removal of residual extract)
  - extract a second time under more severe conditions
    - weekend instead of overnight
    - at 60 °C instead of at ambient temperature
- If second extract contains significant amounts: extraction might be still far from complete
  - swell sample, e.g. in hot water
  - extract by procedure with ethanol (see below)

Extraction of wet foods

- Water is a perfect barrier against extraction with hexane!
  - fills pores, prevents hexane from extracting content in the pores
  - cooked noodles thoroughly mixed into hexane: extraction yield <5 %
- water cannot be evaporated (loss of volatile hydrocarbons)
- sodium sulfate (e.g. fish)
  - homogenized food + 2-4 times amount sodium sulfate (→ no lumps)
  - immersion in hexane overnight
- preferred method: replacement of water by ethanol
  - hexane/ethanol are miscible >10 % water
  - 5 g food + 25 ml ethanol, blending
  - allow to stand for 1 h (exchange with water → pores filled with >80 % ethanol, which provides sufficient solubility)
  - ethanol decanted, hexane overnight/RT
  - combine hexane with ethanol, extract ethanol with water

Extraction of paper and board



- Extraction with hexane or MTBE loads the precolumn with involatile material
  - hotmelts/adhesives
  - binder resins

→ **selective extraction discriminating above <C24**

- Ethanol
  - discriminates against high molecular mass hydrocarbons, particularly n-alkanes
  - improves extraction of <C24 (swelling?)

Extraction yield: second extracts

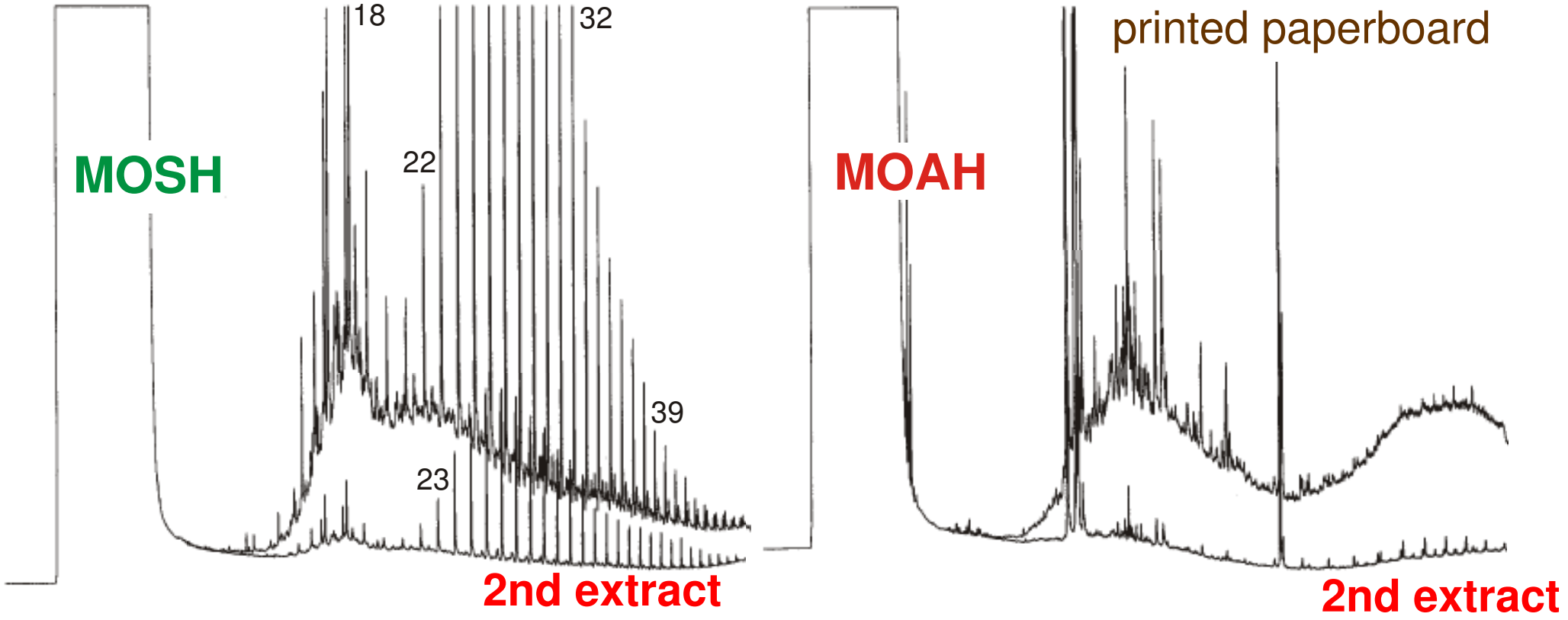
Hexane:ethanol 1:1

2 h/RT sufficient of MOSH and MOAH

– not for diisopropyl naphthalene (DIPN)

Check with second extraction under more severe conditions

– over weekend/RT



Extraction of plastics

Limiting step: diffusion to the surface of the plastic

Strongly varying permeability of polymers

- barriers are not permeable → difficult to extract
- migrated hydrocarbons are more easily extracted than those built in

2 h/RT sufficient for polyethylene

Default conditions: overnight/RT

- testing by second extracts necessary for polymers other than polyolefins

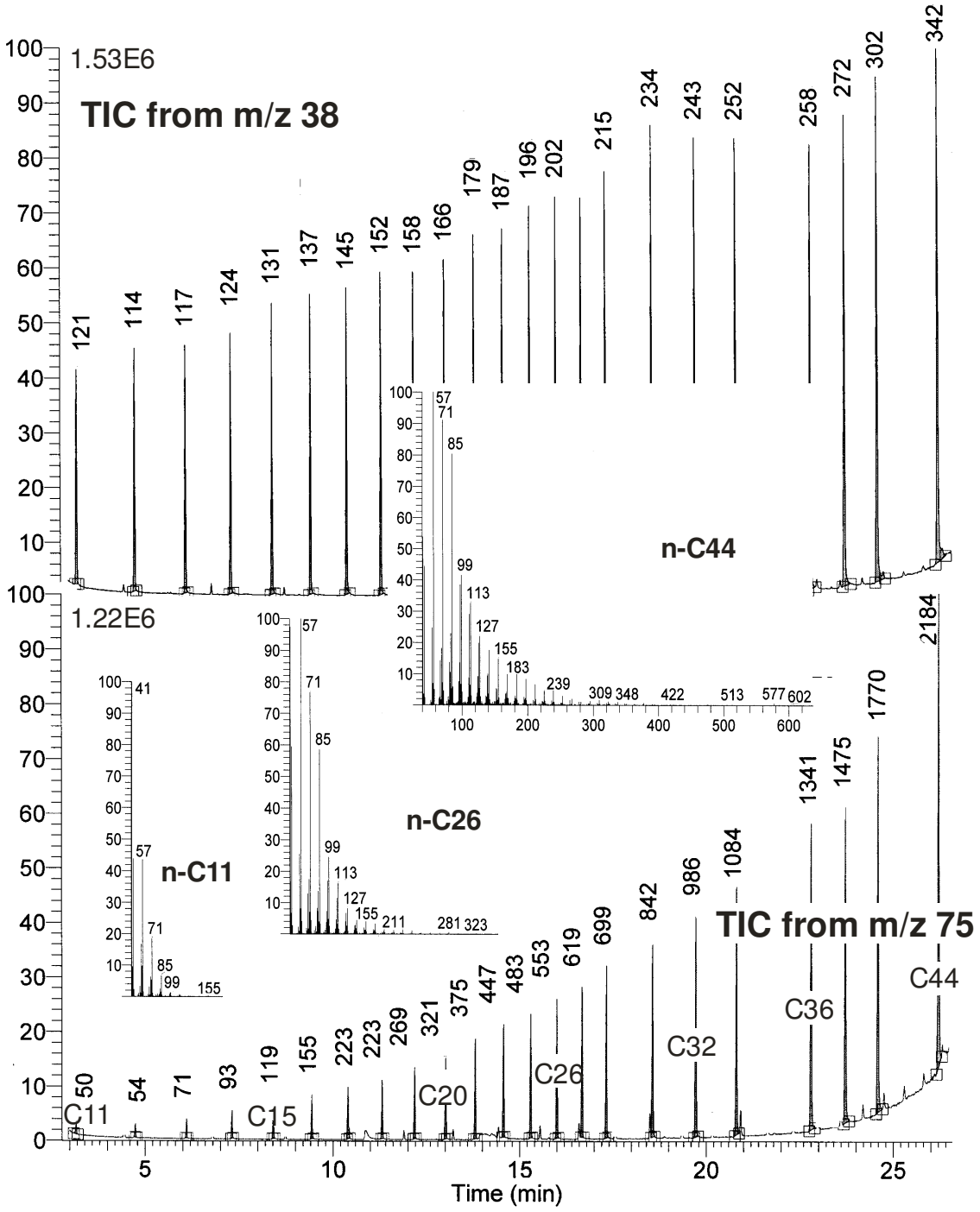
FID as detector

- only detector with equal response for all hydrocarbons (calibration!)
- drawbacks: low sensitivity, no selectivity

→ **enables calibration with any hydrocarbon**

MS in TIC is more sensitive, but response depends on structure. Example: n-alkanes at equal concentrations, EI/TIC on ion trap.

Response increases with mass owing to higher yield of larger fragments (also depending on scanned mass range).



Design of methods: detection limit

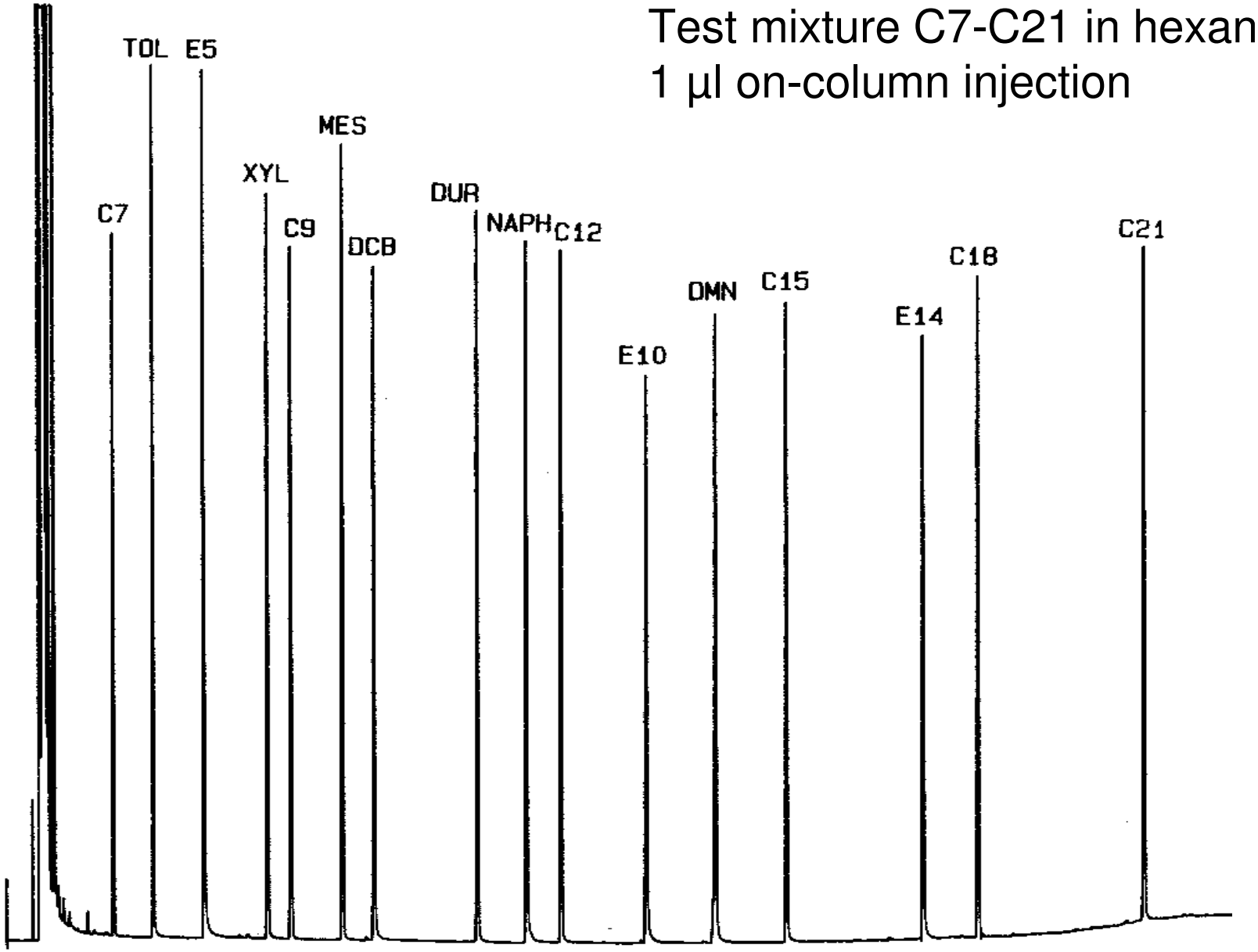
- Quantification limit of FID for a hump of MOSH and MOAH of intermediate width: 50 ng
  >100 times higher than for a clean signal!
- to reach 0.5 mg/kg limit with 50 ng MOSH or MOAH requires aliquot of 100 mg food being injected into GC
- to enable injection of 1 µl, the fractions obtained from the „manual method“ (8-10 ml) would have to be reconcentrated to 10 µl
  - not feasible for routine method
  - loss of volatile components

→ injection of 50 µl is a prerequisite

→ reconcentration to about 500 µl

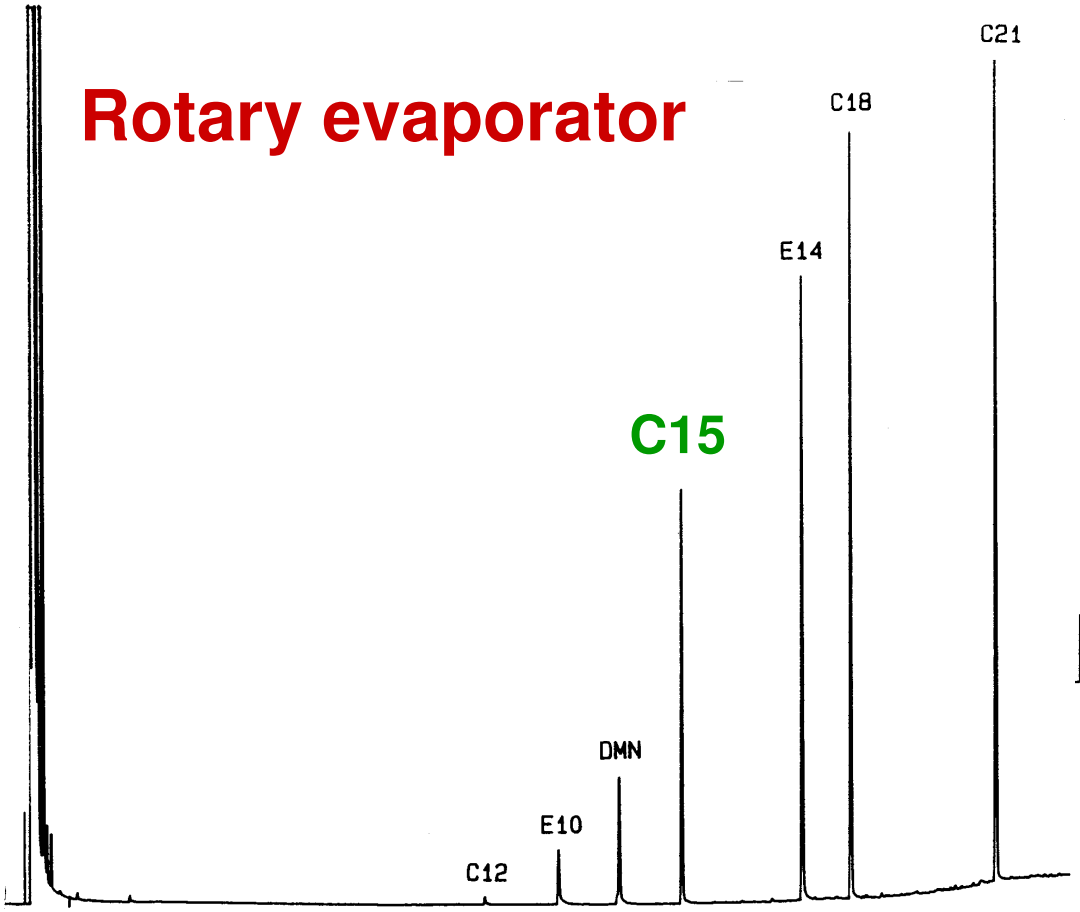
Loss of volatile components

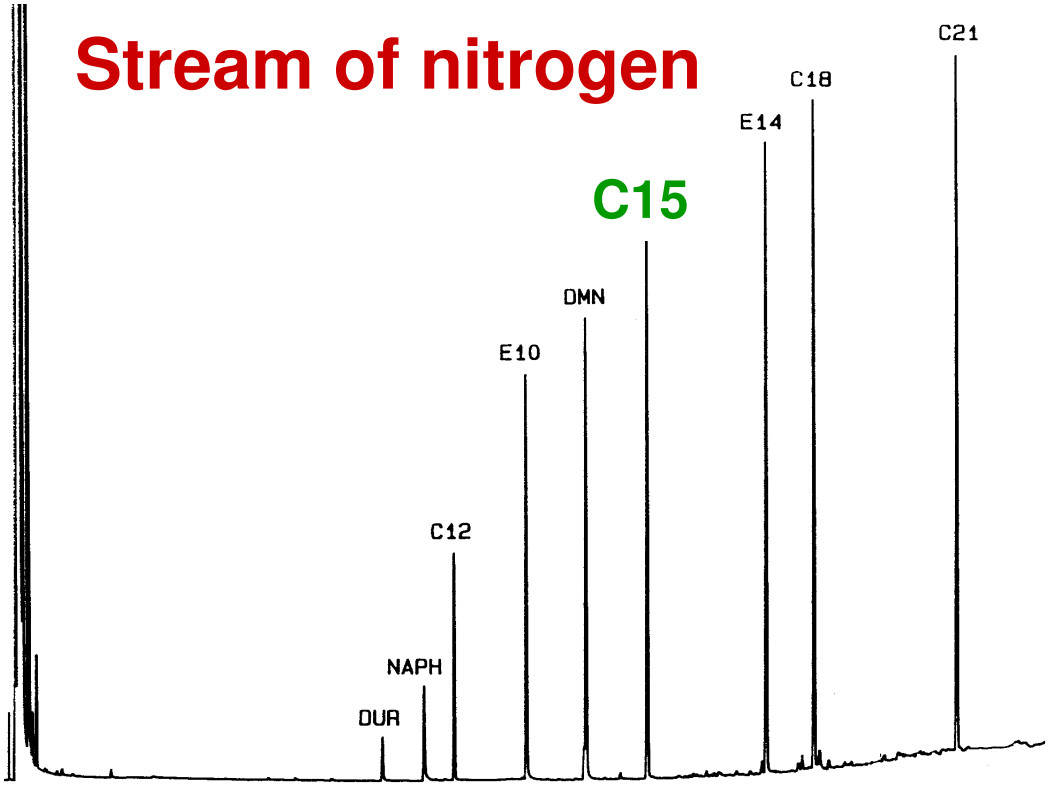
Test mixture C7-C21 in hexane,
1 µl on-column injection



Sample Reconcentration by Column-External Solvent Evaporation or Injection of Large Volumes into GC Capillary Columns. K. Grob and E. Müller. Chromatogr. 404 (1987) 297-305

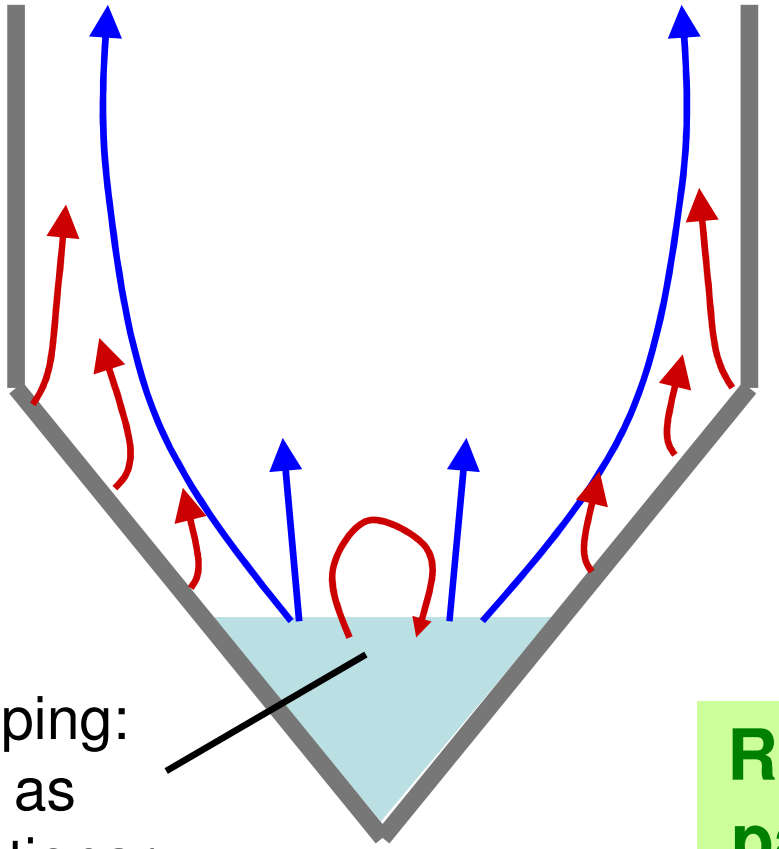
Dilution and reconcentration by a factor of ~80





Often stream of gas is worse than rotary evaporator

Chromatography on the glass wall: volatility depends on the retention power of the wall (material acting as retaining stationary phase)



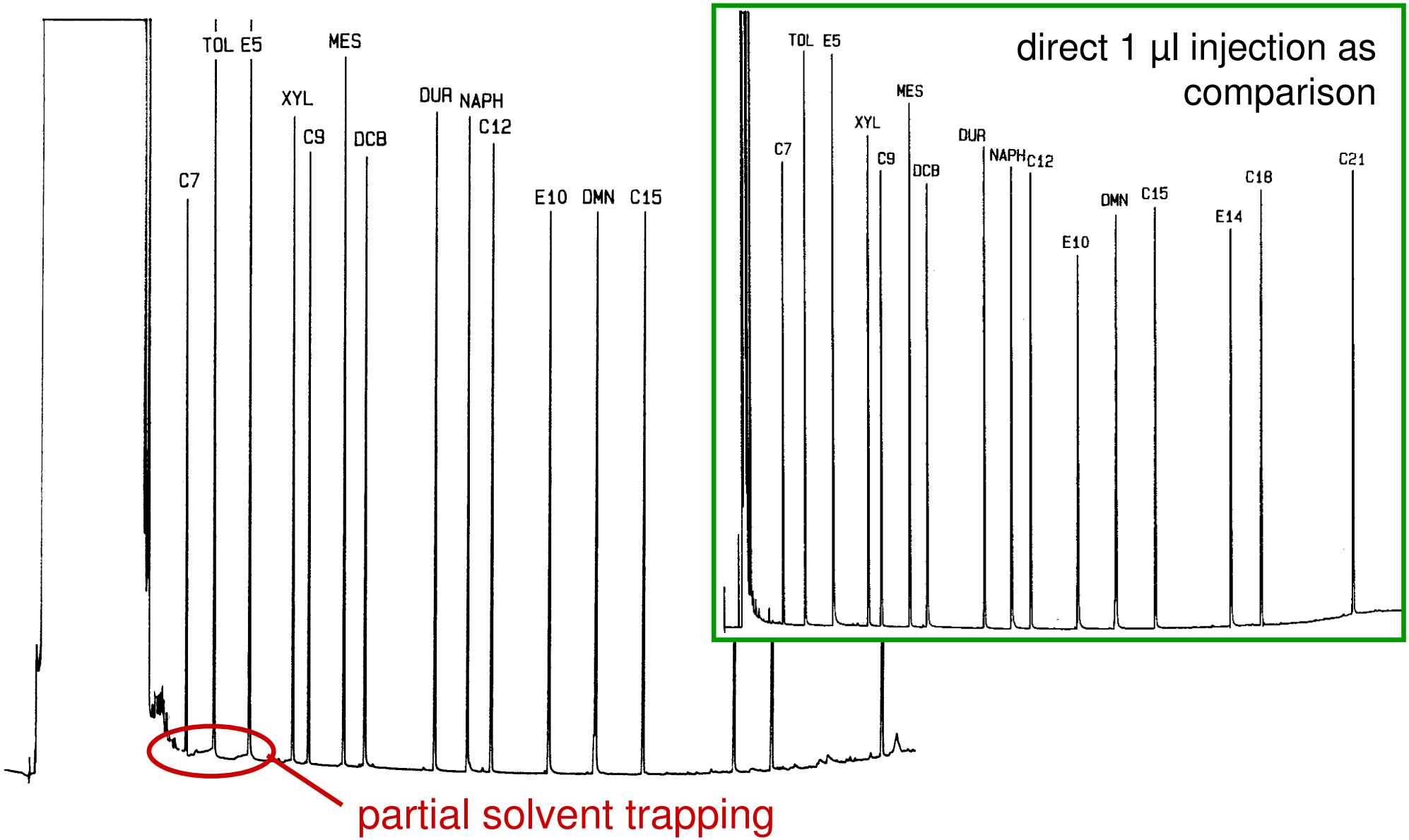
Stream of gas and solvent vapors removes vapors of components

High vapor pressure over dry glass wall (no stationary phase)

Solvent trapping: solvent acts as retaining stationary phase

Reconcentration is particularly critical after preparation!

80 µl injection instead of reconcentration: practically no losses!



Keeper

- Loss of volatiles can be reduced by adding a substance acting as retaining stationary phase
  - such as toluene (manual method)
- Toluene also strongly slows solvent evaporation, rendering the end of solvent evaporation more robust

Capacity to retain lipids

- Fat (primarily triglycerides) removed by retention on LC column
- Triglycerides may flood up to half of LC column, leaving the other half for the isolation of the MOSH and MOAH-fraction
- Capacity of the proposed columns
  - „manual method“: 200 mg
  - HPLC: 20 mg
- Sample extracts can be reconcentrated before preseparation to the limit determined by their fat content
- Limit of quantitation is determined by
  - fat content
  - capacity of the LC preseparation

Limit of quantification

Reconcentration before LC preseparation by fat content:

- low fat (≤4 %) samples (rice, corn, noodles)
  - 10 times → quantitation limit of about 0.1 mg/kg
- medium fat (~20 %) samples (cereals, muesli, biscuits)
  - no reconcentration → quantitation limit of about 0.5 mg/kg
- high fat (~40 %) samples (chocolate)
  - only half amount/concentration → quantitation limit of about 1 mg/kg

Limit of quantification of 0.6 mg/kg not reached for all samples

→ compromise

- LC capacity not adjusted to high fat content
- enrichment on larger column before standard method
- often also removal of plant n-alkanes needed

Tasks for gas chromatography (GC)

- Distinction between mineral and plant hydrocarbons
- Recognition of other interfering components (e.g. DIPN, wood oil components in MOAH)
- Characterization of MOSH and MOAH → source, mode of transfer)
  - molecular mass distribution
    - carbon number at center of elution
    - range of hydrocarbons
  - presence or absence of n-alkanes
- for paperboard: separation at n-C24 (migrating part)
- easy analysis up to about C50