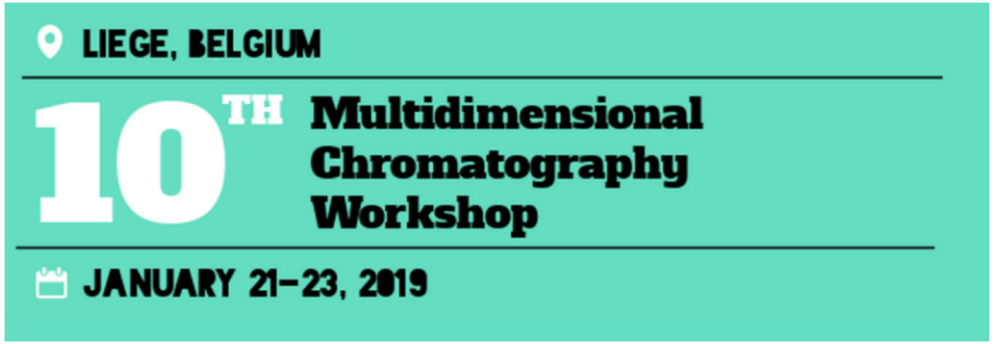
FG3:

Publishing GC×GC data: What are we doing well and what can we improve?

Moderators: Jef Focant & Giorgia Purcaro

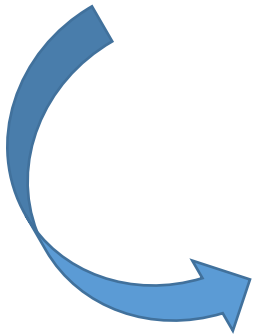


Metabolomics (2007) 3:175–178
DOI 10.1007/s11306-007-0070-6

BRIEF REPORT

The metabolomics standards initiative (MSI)

Oliver Fiehn · Don Robertson · Jules Griffin ·
Mariet van der Werf · Basil Nikolau · Norman Morrison ·
Lloyd W. Sumner · Roy Goodacre · Nigel W. Hardy ·
Chris Taylor · Jennifer Fostel · Bruce Kristal ·
Rima Kaddurah-Daouk · Pedro Mendes ·
Ben van Ommen · John C. Lindon · Susanna-Assunta Sansone



Metabolomics. 2007 September ; 3(3): 211–221. doi:10.1007/s11306-007-0082-2.

Proposed minimum reporting standards for chemical analysis Chemical Analysis Working Group (CAWG) Metabolomics Standards Initiative (MSI)

Metabolomics (2007) 3:175–178
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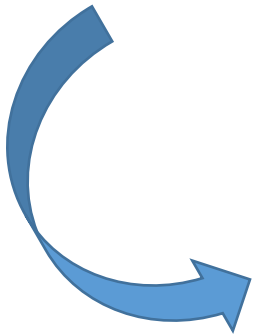
BRIEF REPORT

The Mulitidimensional Standard Initiative

Oliver Fiehn · Don Robertson · Jules Griffin ·
M
L
Chris Taylor · Jennifer Fostel · Bruce Kristal ·
Rima Kaddurah-Daouk · Pedro Mendes ·
Ben van Ommen · John C. Lindon · Susanna-Assunta Sansone

The 10th Multidimensional Workshop



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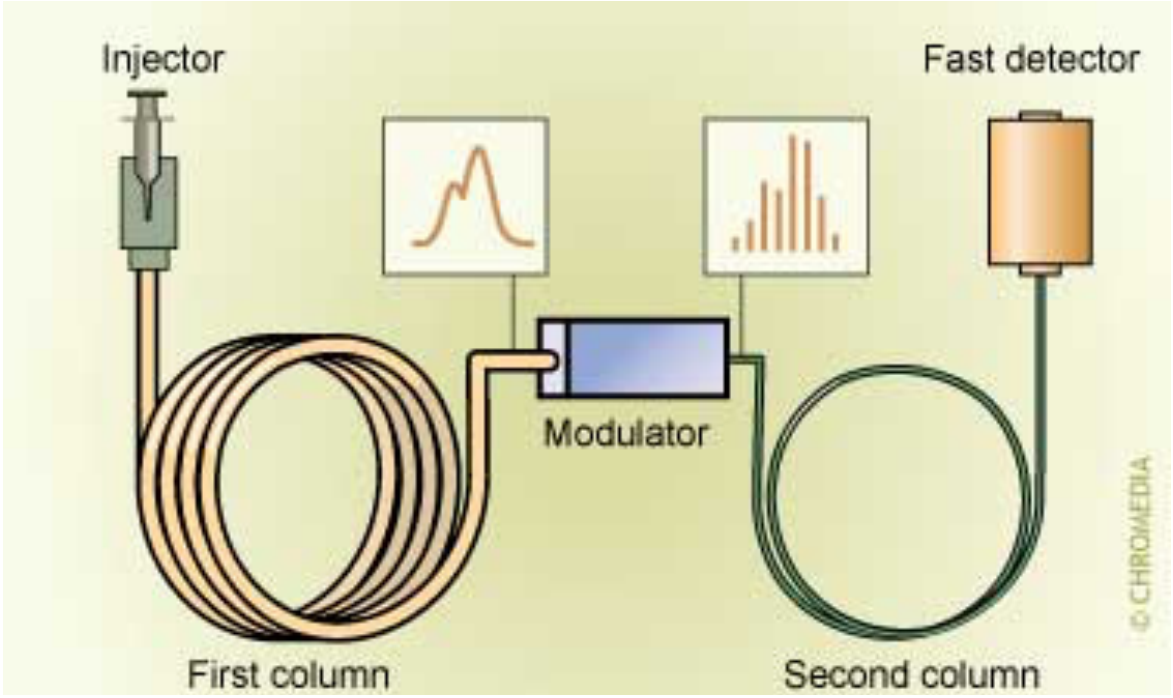
Proposed minimum reporting standards for GC×GC analysis

Chemical Analysis Working Group (CAWG) Metabolomics Standards Initiative (MSI)

Topic 1:

GC×GC Instrumental parameters

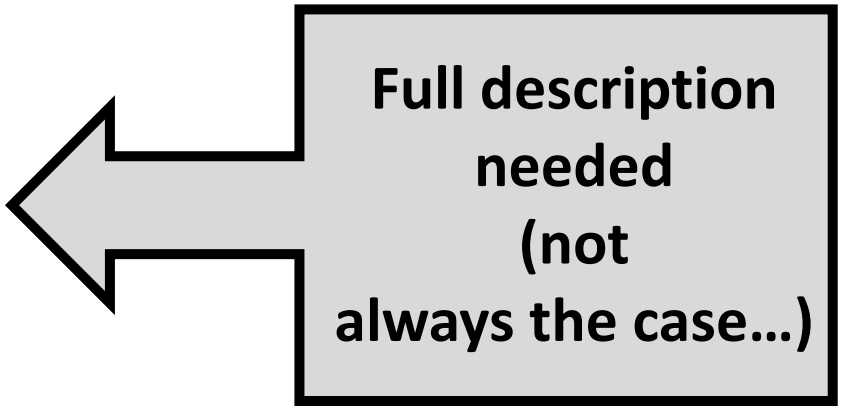


1) Injection parameters?

1a) Hardware information

1b) Software parameters

- Liquid
- HS (SPME, SHS, DHS, TD)



2) GC×GC parameters

2a) Hardware information

- Modulator, connections, …
- Columns

2b) GC×GC parameters

- Oven temperature
- Flow, pressure or linear velocity?
- Modulator:
  - o **Cryogenic**: Modulation time, P_M, hot/cold jet time?
  - o **Flow**: differential vs diverting, auxiliary pressure, flow in the modulator, accumulation time and washing time? Split ratio?
  - o **Phase ratio**: what to mention?
  - o Modulator offset for 'modified' display of chromatograms? How to deal with wraparound?

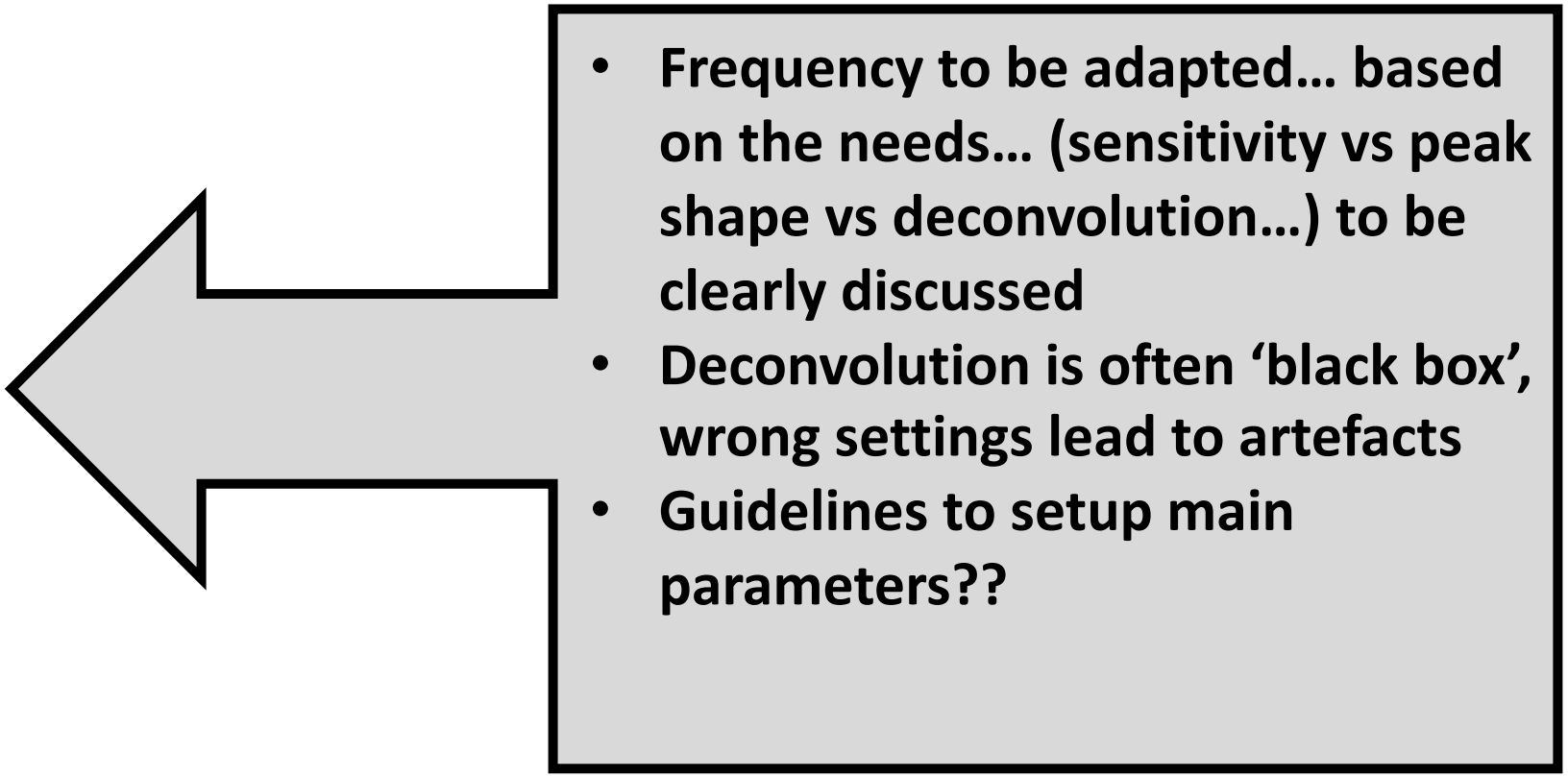
- **Full details to be made available**
- **Refer to previous papers**
- **Jet duration can be important**
- **Add all parameters for replication**
- **NO issue with wrap around**

- **All parameters MUST be listed**

3) Detector

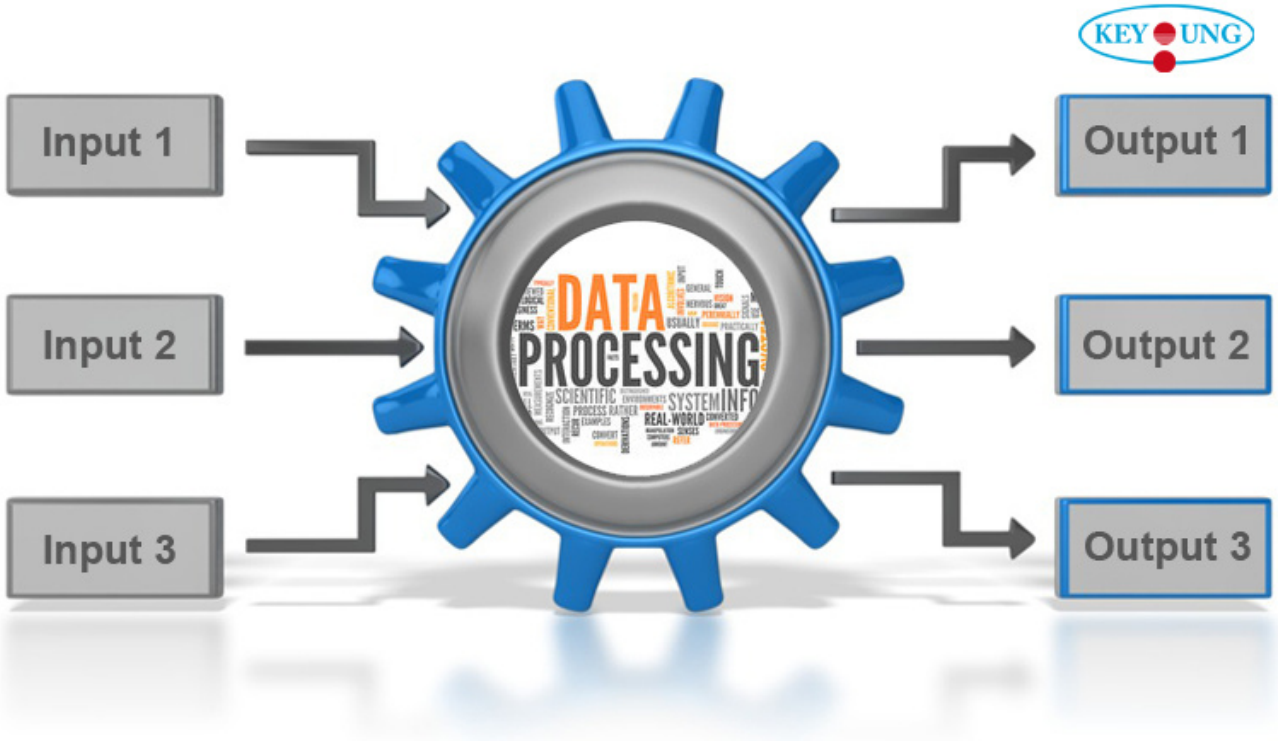
3a) Frequency of acquisition? [try to cover all the main detectors]

- Do we need to state the number of point for peak in average?
- Do we really need 200Hz?
- Deconvolution parameters: what we need to know?



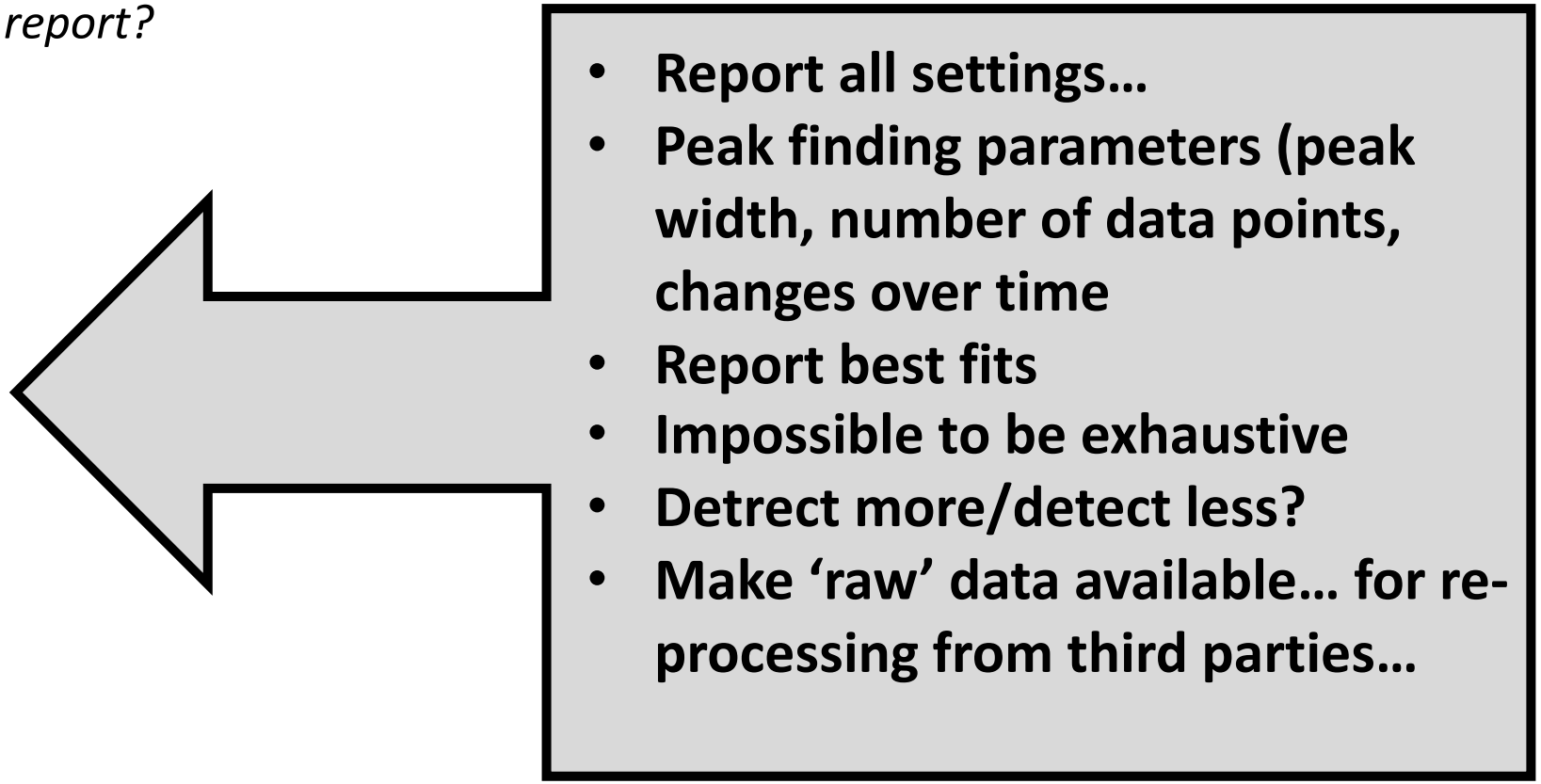
Topic 2:

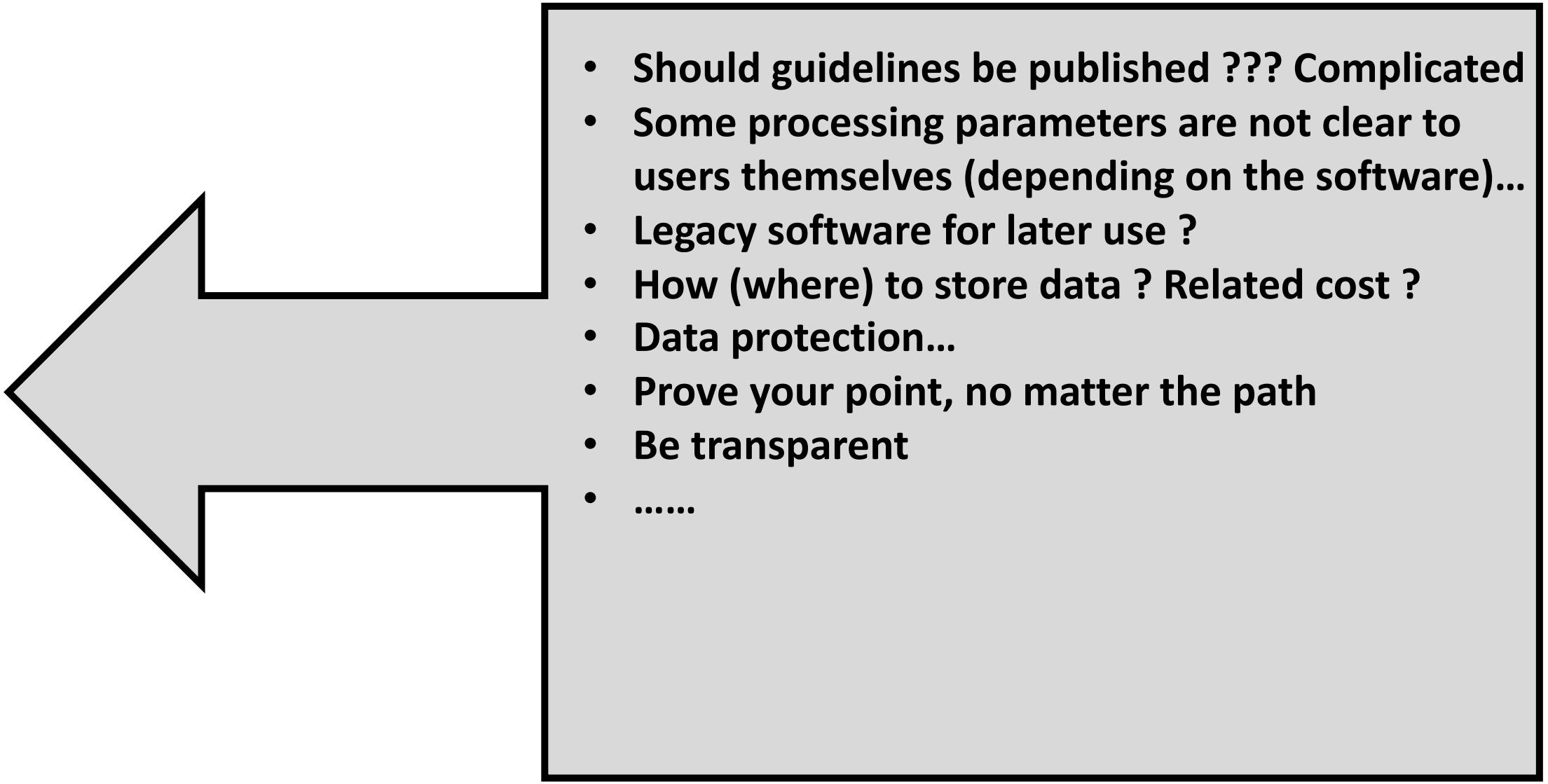
Data processing



2a) Software parameters (Alignment parameters, S/N, Thresholds, etc)

2b) Post processing clean up: what needs to be done manually and what can be automatize? What do we need to report?




Should guidelines be published ??? Complicated
Some processing parameters are not clear to users themselves (depending on the software)…
Legacy software for later use ?
How (where) to store data ? Related cost ?
Data protection…
Prove your point, no matter the path
Be transparent
……

Topic 3:

Quality Control

3a) Tuning results and frequency?

3b) Necessity of (sample, instrumental, …) blanks ?

3c) Standards for instrument check: do we need it?

How the QC-chart should be filled in?

What parameters to be checked? What rules to be applied?

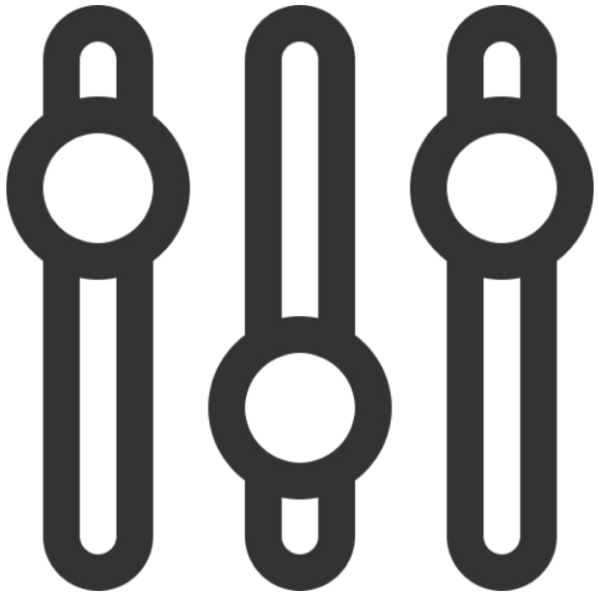
3d) Need for calculation of space occupation/orthogonality?

3e) Should experimental design parameters and plots be included?

- **Rely on instrument tunes…**
- **Demonstrate control of hardware (QC charts on tRs, etc…) MANDATORY in forensics**
- **Space occupation not critical – results are driving methods**
- **Document blanks and report strategy**
- **Include QC samples**

Topic 4:

Do we need a set of different minimum acceptable parameters to be reported according to the field of applications?



IGS™ Scoring Configuration

- [x] Enable Similarity Check

Minimum Similarity for Pass Rating (0 - 999 : 800

Minimum Valid Similarity (0 - 999): 700

- [x] Enable Fragment Ion Check

Minimum Abundance (100 - 998): 300

Required Mass Accuracy: 5 +/- Mass Window ( ) mDa (•) ppm

- [x] Enable Molecular Ion Check

Minimum Library Abundance (0 - 998): 100

Required Mass Accuracy: 5 +/- Mass ( ) mDa (•) ppm

- [x] Enable Retention Index Check

Retention Index Window: 25

4a) What minimum set of parameters for identification ?

-$^{1}t_R$ (LRI, delta LRI ??)

-$^{2}t_R$?

-Library match value ? (forward, reverse, probability, …)

-Mass accuracy ?

-Pure standard injections ?

-…

4b) What about initiatives like “Identification Grading System” ?

-What default value to set ?

- **Difficult to define how confident we have to be**
- **Subjective view points depending on reviewers/journals**
- **Need for a set of defined values**
- **Some proof of concept studies still needed**
- **Use of a ‘scoring system’**

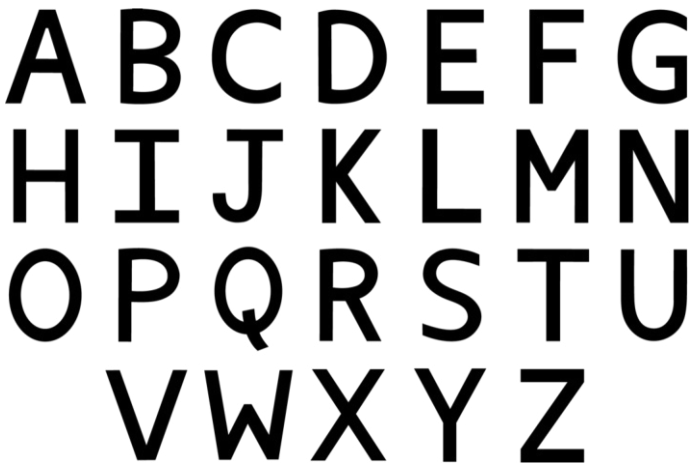
Topic 5:

Should we use the current ‘official nomenclature’?

Is it time for an updated nomenclature?

Do we need a nomenclature?

Column sets nomenclature to be used?



GCxGC Nomenclature

Nomenclature and Conventions in Comprehensive Multidimensional Chromatography

Peter Schoenmakers,[a,b] **Philip Marriott**[c] **and Jan Beens,**[d]
[a]Polymer-Analysis Group, University of Amsterdam, The Netherlands,
[b]Dutch Polymer Institute, Eindhoven, The Netherlands,
[c]Department of Applied Chemistry, RMIT University, Melbourne, Australia,
[d]Faculty of Science, Free University, Amsterdam, The Netherlands.

LC•GC Europe June 2003

Nomenclature and Conventions in Comprehensive Multidimensional Chromatography

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[c]Department of Applied Chemistry, RMIT University, Melbourne, Australia,
[d]Faculty of Science, Free University, Amsterdam, The Netherlands.

LC•GC Europe May 2012

- **Nomenclature is very important**
- **Users MUST use proper terms and layout**
- **People to speak the same language**

Table 3(a): Nomenclature suggested for comprehensive two-dimensional (gas) chromatography.

Term	Definition
Modulator	Interface device between the two columns in a comprehensive two-dimensional separation system that accumulates or samples narrow bands from the eluate of the first column for fast re-injection into the second column.
Modulation time or Modulation period (P_M)	The duration of a complete cycle of modulation in a comprehensive two-dimensional separation system (equals the data conversion time of each second dimension chromatogram, *i.e.*, the time between two successive injections into the second column).
Modulation frequency (f_M)	Number of modulations per unit of time.
Modulator temperature (T_M)	The temperature of the modulation zone used in thermal modulation.
Modulation number (n_M)	The number of modulated peaks recorded for a given first-dimension peak.
Modulation ratio (M_R)	The ratio of the peak width at baseline (^1w_b) for the first dimension peak to the modulation period (P_M).
Modulation phase (Φ; F_M)	The pattern of modulated peaks caused by the time relationship between the shape of the analyte peak and the pulsing process of the modulator in a comprehensive two-dimensional separation system (18).
In-phase modulation	The modulation phase that produces a symmetrical sequence of peaks with a single maximum peak pulse (18).
Out-of-phase modulation	Any phase that produces a non-symmetrical peak-pulse distribution (18).
180° out-of-phase modulation	The modulation phase that produces a symmetrical sequence of peaks with two equal maximum peak pulses (18).
Single-stage modulation	Accumulation and focusing during one series of processes at one location in the modulator.
Dual-stage modulation	Accumulation and focusing during two successive series of processes at two locations in the modulator.
Focusing effect	Reduction of the band width (in time, distance and/or volume units) (= band width without modulation/band width with modulation).
Sensitivity enhancement (= peak-amplitude enhancement)	Ratio between peak height with and without modulation (note: sensitivity refers to the signal, not to the noise!).*
Zone compression	The effect of reducing a chromatographic peak (width) in space or time to give a higher concentration within a chromatography column.

Table 3: Nomenclature suggested for comprehensive two-dimensional (gas) chromatography.

Separation space	The region within the two-dimensional GC×GC plot in which compounds are, or may be, distributed.
Wrap-around	The occurrence of second dimension peaks in subsequent modulation sequences, caused by second-dimension retention times that exceed the modulation period of a comprehensive two-dimensional system (19).
Iso-volatility curves	The observation of reduced retention of a solute on a ^{2}D column in GC×GC as the temperature of the oven increases, seen as a decreasing retention time ^2t_R band in the 2D plot.
Column set	The combination of columns used for a given comprehensive 2D chromatography experiment.
Column set relative diameter ratio	The relative change in cross sectional area for the ^{1}D to ^{2}D columns of the column set = $^1d_c/^2d_c$.
Chromatogram structure	The observed ordering of chemically related compounds in the plane of a comprehensive two-dimensional separation.
Colour plot	Two-dimensional plot representing a comprehensive two-dimensional separation, in which the colour represents the signal intensity of the components in the separation system.**
Contour plot	Two-dimensional plot representing a comprehensive two-dimensional separation, in which similar signal intensities of components are connected by means of a line.**
Apex plot	Two-dimensional plot representing a comprehensive two-dimensional separation, in which peak apexes of second-dimension peaks are displayed by a symbol in the ^{2}D space. This may also be simplified to the peak apexes of individual components. **
Cryogenic / thermal modulation	GC×GC system in which the interface operates by changes in temperature compared with the oven temperature, either by setting an elevated or cooler temperature.
Diaphragm modulation	GC×GC system in which the interface operates by periodically selecting a small sub-fraction of the ^{1}D peak to be transferred to the ^{2}D column using a diaphragm system.
Flow modulation	GC×GC system in which the interface operates by a flow switching mechanism; normally a higher flow is maintained for the ^{2}D column.

* A reduction in the detection limit may also be achieved. This reduction is proportional to the product of the sensitivity enhancement and the noise.

** The x-axis represents the first-dimension retention time, the y-axis the second-dimension retention time of the separation system.

Table 4: Non-exhaustive list of symbols suggested for use in GC×GC (and other comprehensive 2D separation methods).

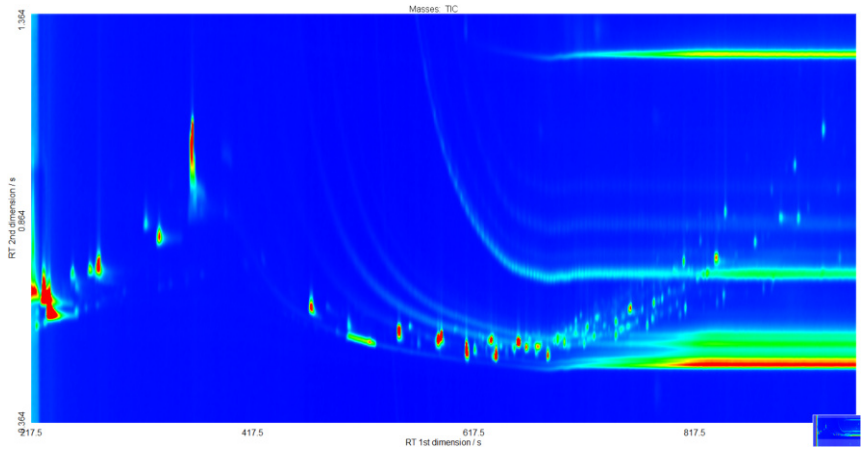
Symbols	Definition
${}^1d_c$, ${}^2d_c$	Internal diameters of the first- and second-dimension columns (respectively) used in a comprehensive two-dimensional system.
${}^1D$, ${}^2D$	First dimension and second dimension of a C2DC system
1D, 2D	One dimensional or two-dimensional system
${}^1t_R$, ${}^2t_R$	Retention times of a peak in the first and second dimension of a comprehensive two-dimensional system (respectively). Note that ${}^2t_R$ can potentially differ for each modulated peak of a given injected component.
${}^1t_M$, ${}^2t_M$	Hold-up times (or "dead" times) of the first and second columns of a comprehensive two-dimensional system (respectively).
${}^1k$, ${}^2k$	Retention factors of a peak eluting from the first-and second-dimension columns of a comprehensive two-dimensional system (respectively)
${}^1I$, ${}^2I$	Retention indices of a peak eluting from the first- and second-dimension columns of a comprehensive two-dimensional system (respectively)
${}^1N$, ${}^2N$	The numbers of theoretical plates of the first and second columns of a comprehensive two-dimensional system (respectively).
${}^1N_{eff}$, ${}^2N_{eff}$	The numbers of effective plates of the first and second columns of a comprehensive two-dimensional system (respectively).
${}^1\sigma$, ${}^2\sigma$	Standard deviations of a peak eluting from the first-and second-dimension columns of a comprehensive two-dimensional system (respectively).
${}^1w_b$, ${}^2w_b$	Peak widths at base of a peak eluting from the first-and second-dimension columns of a comprehensive two-dimensional system (respectively).
${}^1R_s$, ${}^2R_s$	Resolution values of a peak pair eluting from the first and second column of a comprehensive two-dimensional system (respectively).
${}^1n_c$, ${}^2n_c$	Peak capacities of the first and second columns of a comprehensive two-dimensional system (respectively) [the use of n_c is advised to avoid confusion with n that is sometimes used for theoretical plates]
${}^1d_f$, ${}^2d_f$	Film thicknesses of the first and second columns of a comprehensive two-dimensional system (respectively).
${}^1\bar{\mu}$, ${}^2\bar{\mu}$	Average linear velocities in the first and second columns of a comprehensive two-dimensional system (respectively).
${}^1T_e$, ${}^2T_e$	Elution temperatures for a peak eluting from the first dimension and second dimension of a comprehensive two-dimensional GC system (respectively). (note that ${}^1T_e$ and ${}^2T_e$ will be essentially the same due to the very fast elution of components on ${}^2D$ for the GC×GC experiment, defining isothermal elution)
T_M	Modulator temperature
P_M	Modulation period
M_R	Modulation ratio
${}^1t_{R,app}$	Apparent first dimension retention time of the component on the first dimension
t_h	Hold time of the peak in the modulator
As_{2D}	Two-dimensional peak asymmetry

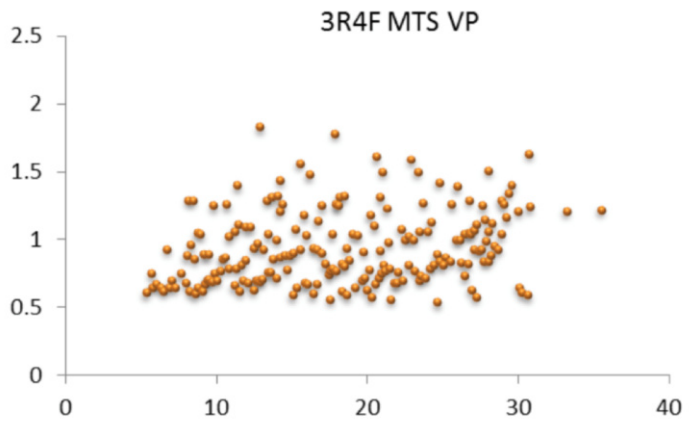
Topic 6:

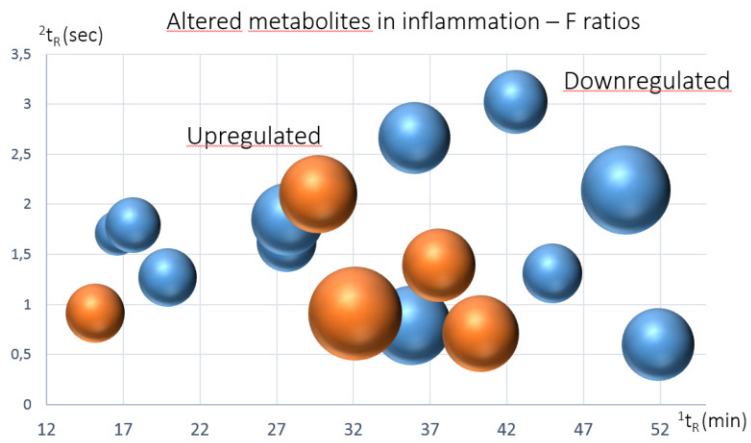
Do we need to always show at least one chromatogram?

What about apex plots *vs* real peaks?

Pseudochromatograms ? FR ‘bubble’ plots?







- Apex plots are an efficient way to report chromatographic data
- Always support findings with chromatograms
- ‘Bubble plots’ efficient to localize relevant analytes
- Bubbles can hide other analytes if too big…
- Use a mix of displays to illustrate and support findings

Topic 7:

What should Supplementary Materials typically made of?

How detailed should they be?

Should raw data be made available?

Untargeted Blood Metabolic Profiling by GC×GC-HRTOF-MS

Supporting Information

Nicolas Di Giovanni[†], Marie-Alice Meuwis[‡], Edouard Louis[‡], Jean-François Focant[*†]

Affiliation:

[†]University of Liège, Department of Chemistry, Organic & Biological Analytical Chemistry Group, Allée du 6 août, B6c, B-4000 Liège (Sart Tilman), Belgium

[‡] University of Liège, GIGA institute, Translational gastroenterology, Uliège, and CHU de Liège, Hepato-gastroenterology and digestive Oncology, Quartier Hôpital, Avenue de l'hôpital 13, B34-35, B-4000 Liège, Belgium.

CONTENTS

[*]To whom correspondance should be addressed:

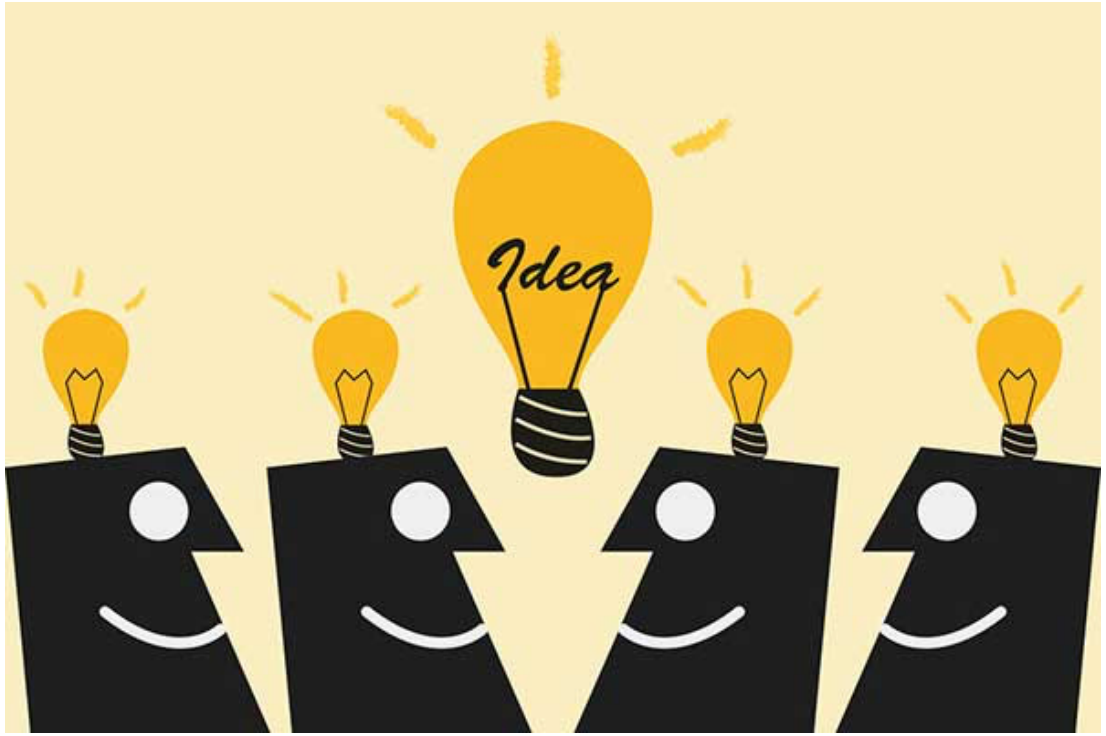
Phone: +32 366 35 31, Fax: +32 4 366 43 87, E-mail: jf.focant@uliege.ac.be

- **How much is too much ?**
- **What is reasonable ?**

84 pages….

Topic 8:

What else?




Thank You