

Heated tobacco product aerosol generation and analysis by gas chromatography

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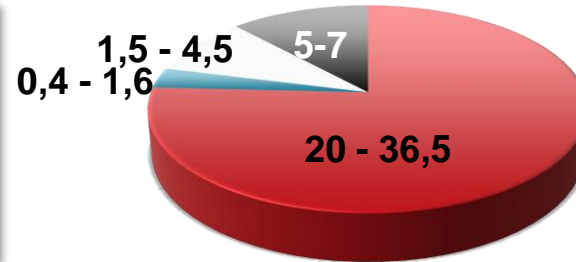
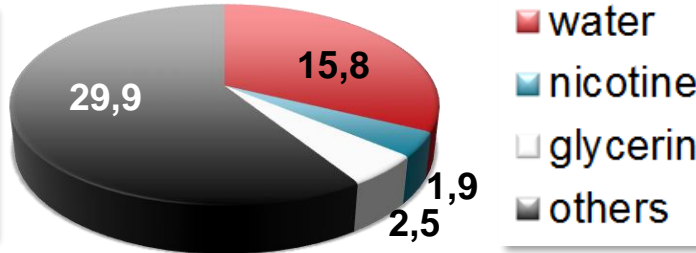
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1. Context

Burning cigarette smoke
Total Particulate Matter (TPM)**
mg/cig

Heated Tobacco Product aerosol
Aerosol condensed matter (ACM)**
mg/stick



** HCl smoking regime

Composition of HTP aerosol is different from a burning cigarette

1. Context

Reference methods for cigarettes ISO 3308

	Puff duration	Puff interval	Puff volume
ISO*	2	60	35
HCI**	2	30	55

* International standardisation organisation

** Health Canada intense smoking regime with blocked vents

No smoking reference methods defined for HTP



Design	IQOS [®]	glo [™]	Ploom S [°]
Heating period *** (s)	340	180 (G002) 210 (G004)	210
Pre heating (s)	20	40	40
Max puffs -ISO	6	4	4
Max puffs-HCI	12	7/8	8

***Heating period for puff generation without pre heating

°Device limited to 14 puffs whatever the puff interval

1. Objectives

Develop a method :

- to generate and collect HTP aerosol
- to separate nicotine, humectants and water by Gas Chromatography (GC) and quantify them with Flame Ionization Detection (FID) and Thermal Conductivity Detection (TCD)

Assess how the HTP stick conditioning and puff interval affect aerosol composition of various products



2. Methodology : aerosol generation & collection

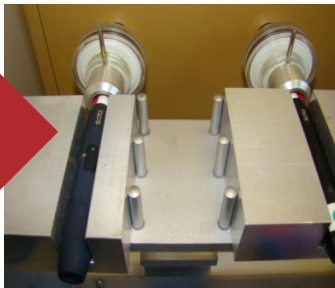
Smoking conditions (ISO3308) : HCI smoking regime, vents unblocked

Linear smoking machine



Heaters

- Cleaned
- At room temperature
- Fully charged



3 sticks heated per filter holder
(with 3 heaters)

Puffing starting at the end of pre-heating

Filter holder



ACM collection on a glass fiber filter pad (\varnothing 43 mm)

Closed with caps immediately after smoking
ACM weight measurement

2. Methodology : aerosol extraction

Filter pad Ø 43 mm with ACM



Condensed matter on filter holder parts recovered with 2 x 1/4 filter pad



Add methanol (20 ml) with internal standards

- n-heptadecane (0,5 g/l)
- isopropanol (10 ml/l)



Dried 100 ml flask

Flask shaking for 1 h at 350 rpm before vials preparation

2. Methodology : aerosol analysis



**Shimadzu 2010 Plus GC
with AOC-20i Dual Tower**

GC with 2 columns and detectors

	FID	TCD
Analytes	PG, nicotine, glycerin	Water
Internal standard	n-heptadecane	isopropanol
Injection volume	1 μ l	1 μ l
Oven parameters	170-245°C gradient	
detector T°C	270 °C	250 °C
Total run time	11 min	

2. Methodology : calibration

Analyte	Standards number	Range mg/ml	r ²	Calibration model	LOD* mg/ml	LOQ* mg/ml
Nicotine	7	0-1	0,9991	aX	0,008	0,023
Glycerin					0,012	0,036
PG					0,004	0,013
Water	7	0-4	0,9993	aX+b	0,035	0,118

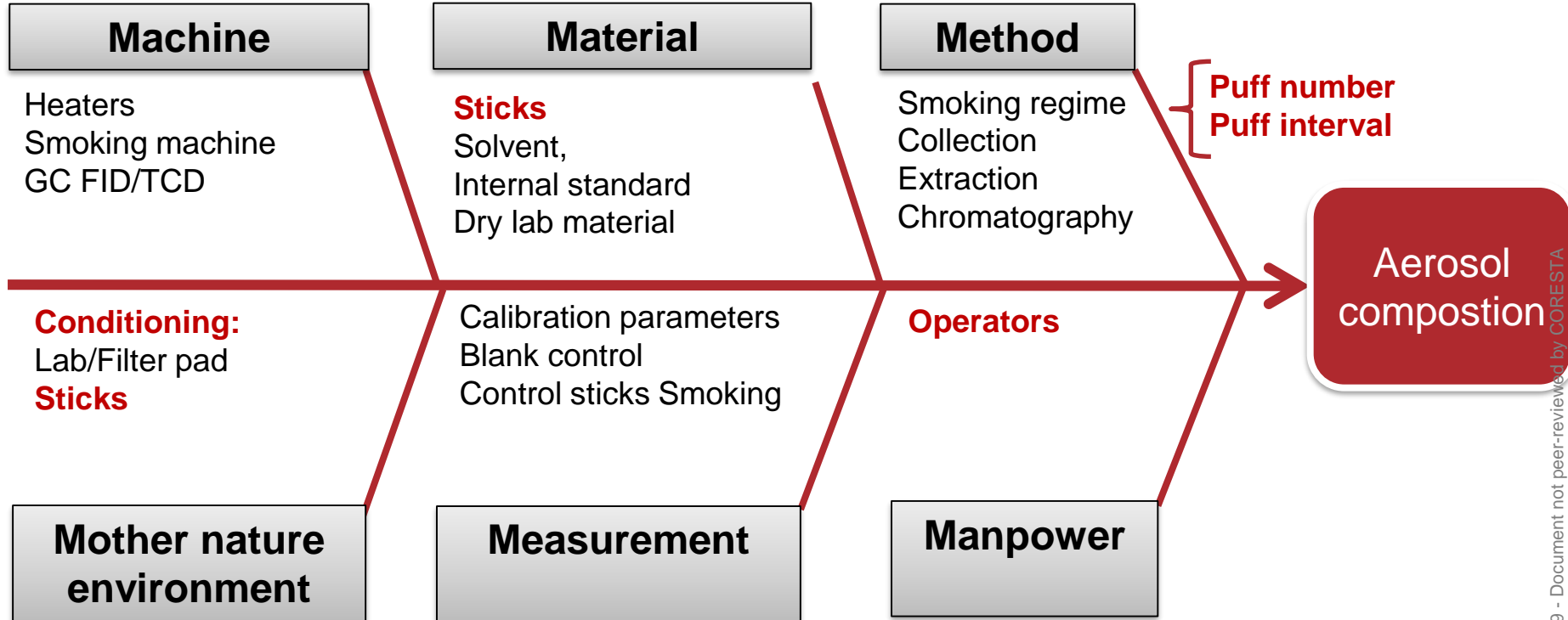
* Calculated according to ISO 5725

Calibration standards validity : 2 weeks at 20°C ± 2°C, protected from light

Registered parameters: slope coefficient, internal standard area, retention time

Reference sticks heated at each smoking session and registered on a control chart

2. Methodology: Influent parameters



3. Effect of puff interval

Human smoking topography (clinical studies)

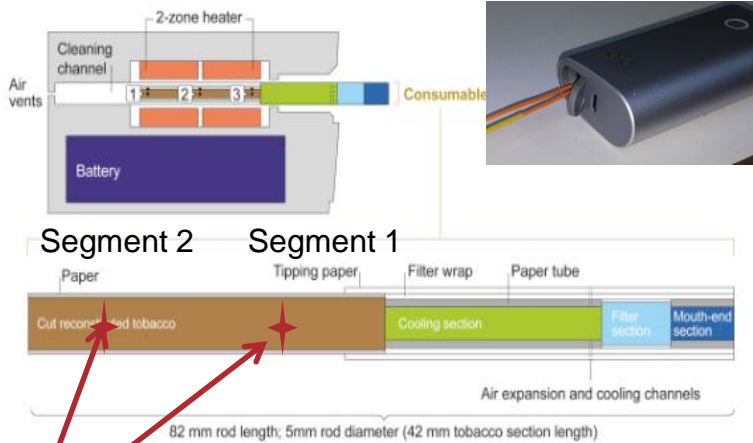
Type of product	Puff number mean range	Puff interval mean \pm SD ² or CI ³ (s)
Heatstick in IQOS [®]	15,5 - 17,7	13,3-15,7 ¹
Neostiks [™] in glo [™]	10,9 - 12,3	7,7 \pm 3,9 ²
Heatstick in IQOS [®]	10,3 - 10,8	8,6 \pm 3,1
Neostiks [™] in glo [™]	11,0 - 12,5	9,4 \pm 3,1 ³
Heatstick in IQOS [®]		10,3 \pm 3,4

¹C. Haziza et al. / Regulatory Toxicology and Pharmacology 81 (2016) 489-499-realculated data vs puff number per min

²J. Gee et al./ Regulatory Toxicology and Pharmacology 93 (2018) 84-91

³J. Murphy et al. / Regulatory Toxicology and Pharmacology 93 (2018) 92-104

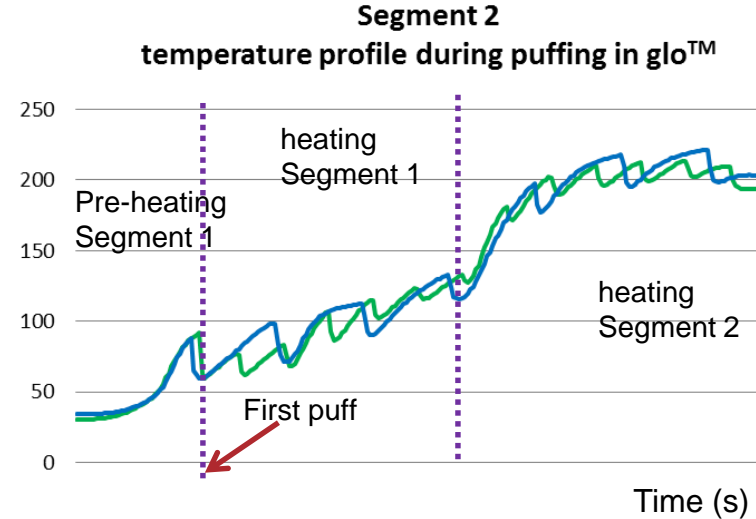
3. Effect of puff interval



Thermocouples in middle of the tobacco rod

Puff interval

- 15 s
- 30 s



15s vs 30s :Variation in temperature profil & amplitude
Potential effect on aerosol composition ?




Schema from
Forster et Al. *Regulatory Toxicology and Pharmacology* (2018) vol: 93 ,14-33

3. Effect of puff interval : methodology

Smoking conditions

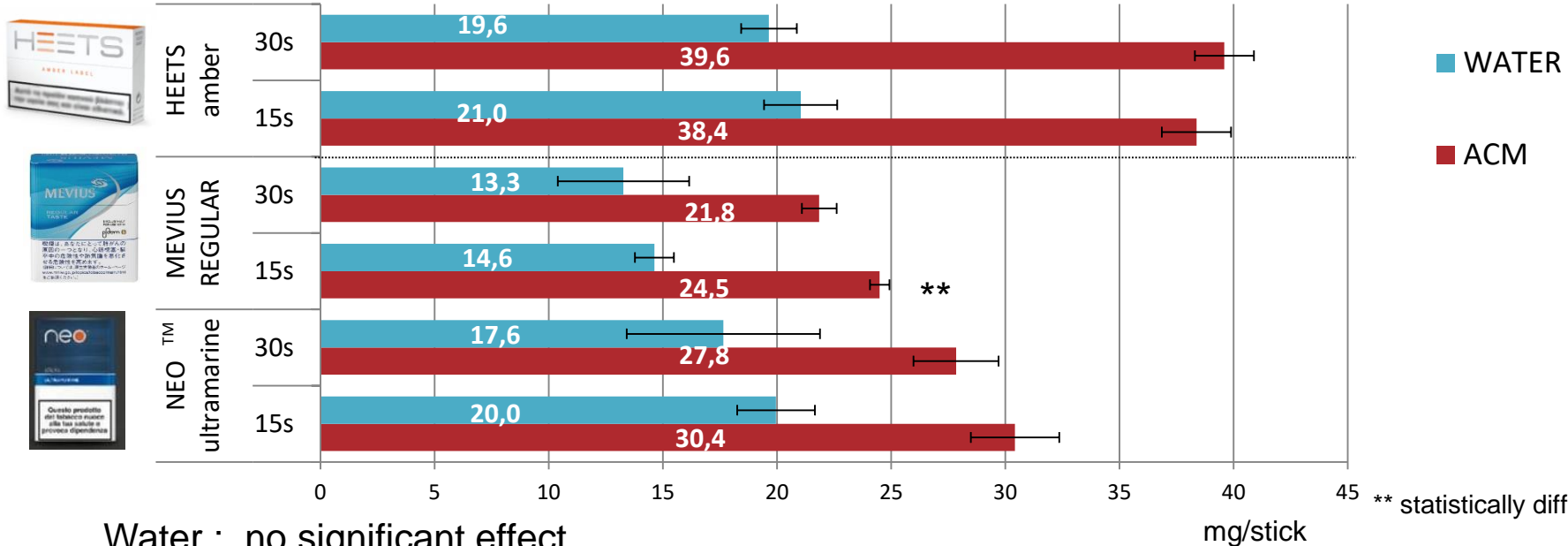
HCI : 15 s & 30 s puff interval, puff duration 2 s, puff volume 55 ml, unblocked vents

Sticks from sealed pack

Heaters	Sticks		Number of replicates	Puff number for 15s puff interval	Puff number for 30s puff interval
IQOS®	HEETS Amber (France)		14	12	12
Ploom S	MEVIUS Regular (Japan)		6	12*	10*
glo™ G004	NEO™ Ultramarine (Italy)		6	12*	10*

3. Effect of puff interval : results

Aerosol composition : ACM & water (mg/stick: mean \pm CI_{95%})

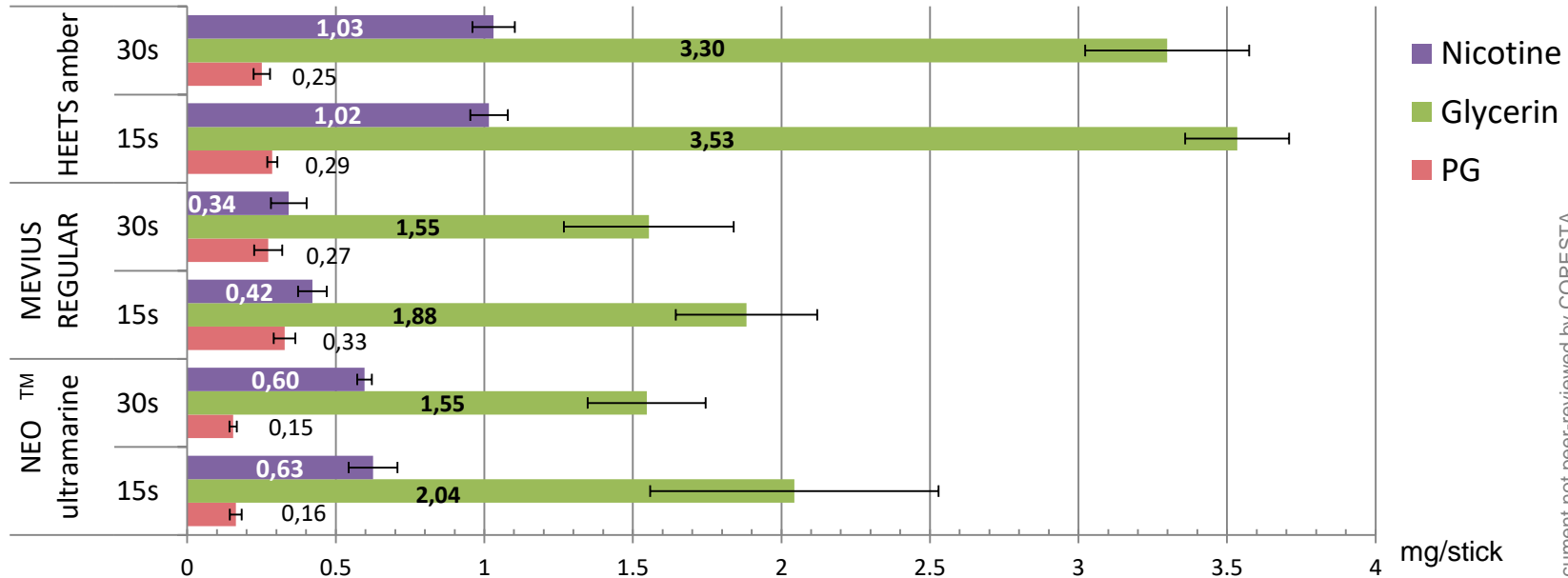


Water : no significant effect

ACM : no significant effect except for Mevius (+12%) (puff number effect)
variation per puff < method relative error

3. Effect of puff interval : results

Aerosol composition : nicotine, glycerin, PG (mg/stick: mean \pm CI_{95%})



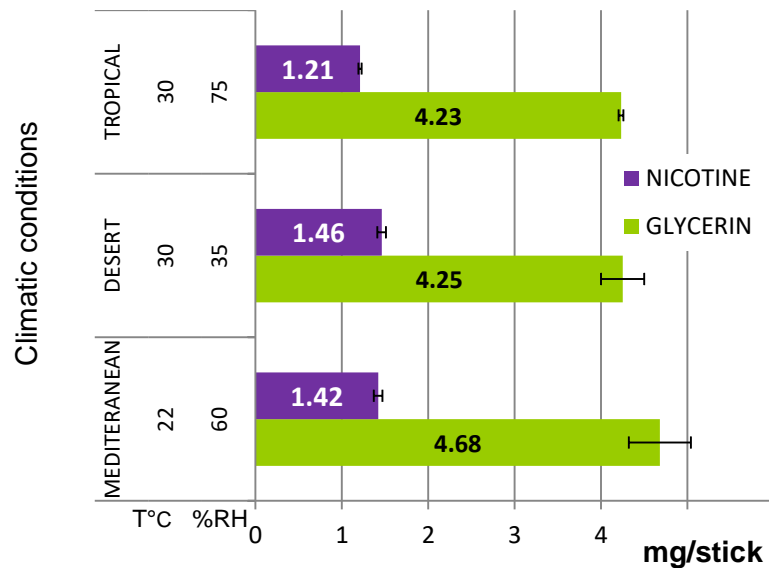
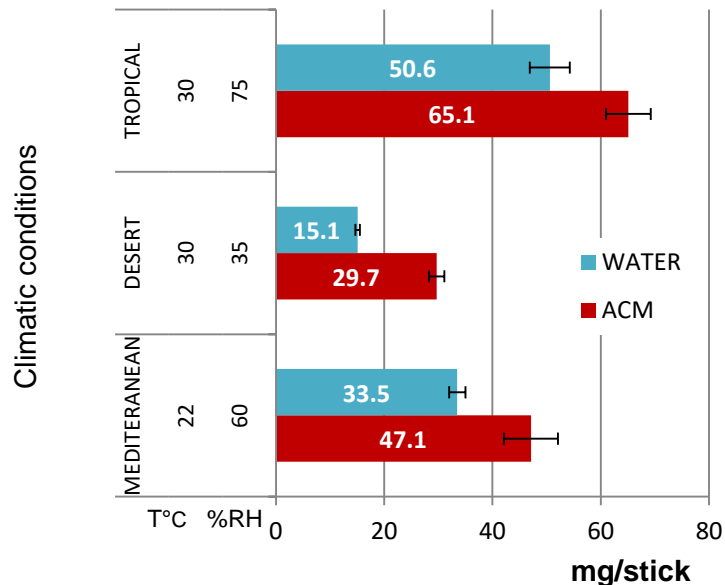
No significant variation for nicotine, glycerin, PG

4. Effect of stick conditioning

Effect of climatic conditions on aerosol composition,

Schaller et al. Regulatory Toxicology and Pharmacology 81 (2016) S27-S47

Heatsticks (THS2.2) in IQOS®, HCl smoking regime, 48h sticks conditioning, mg/stick: mean ± CI_{95%}

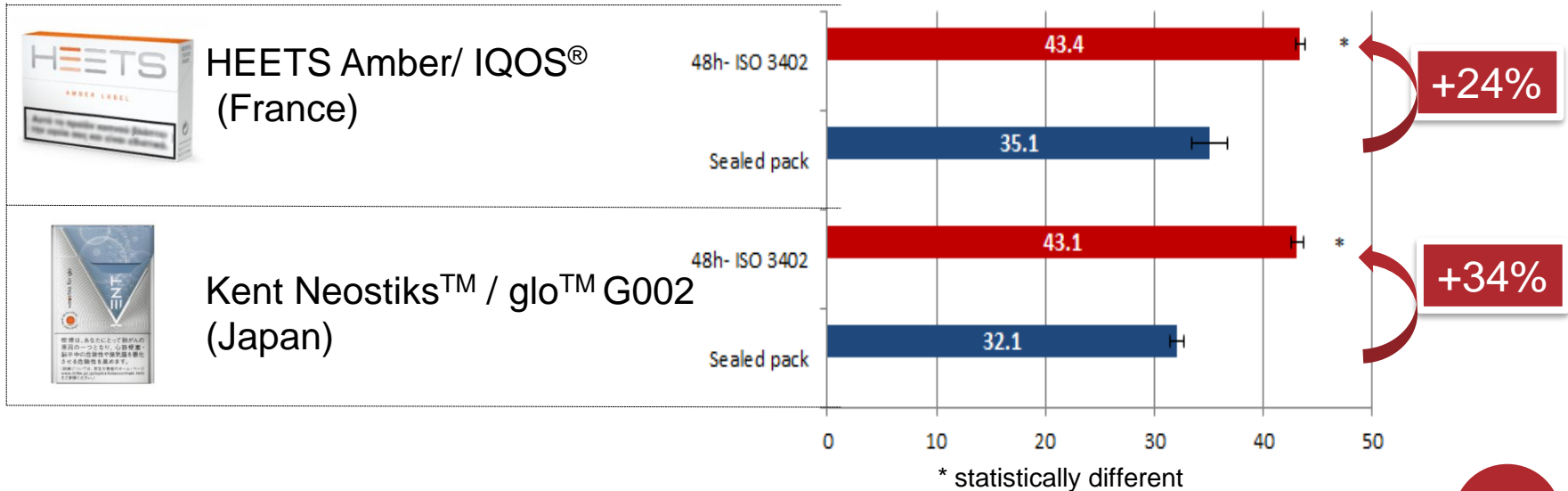


4. Effect of stick conditioning : methodology

■ Sticks 48h at RH=(60 ± 3)%, T =(22 ± 2)°C (ISO 3402)

■ Sticks in sealed pack : 48h at T =(22 ± 2)°C

Tobacco composition after conditioning:
water (GC) mg/stick : mean ± IC_{95%}

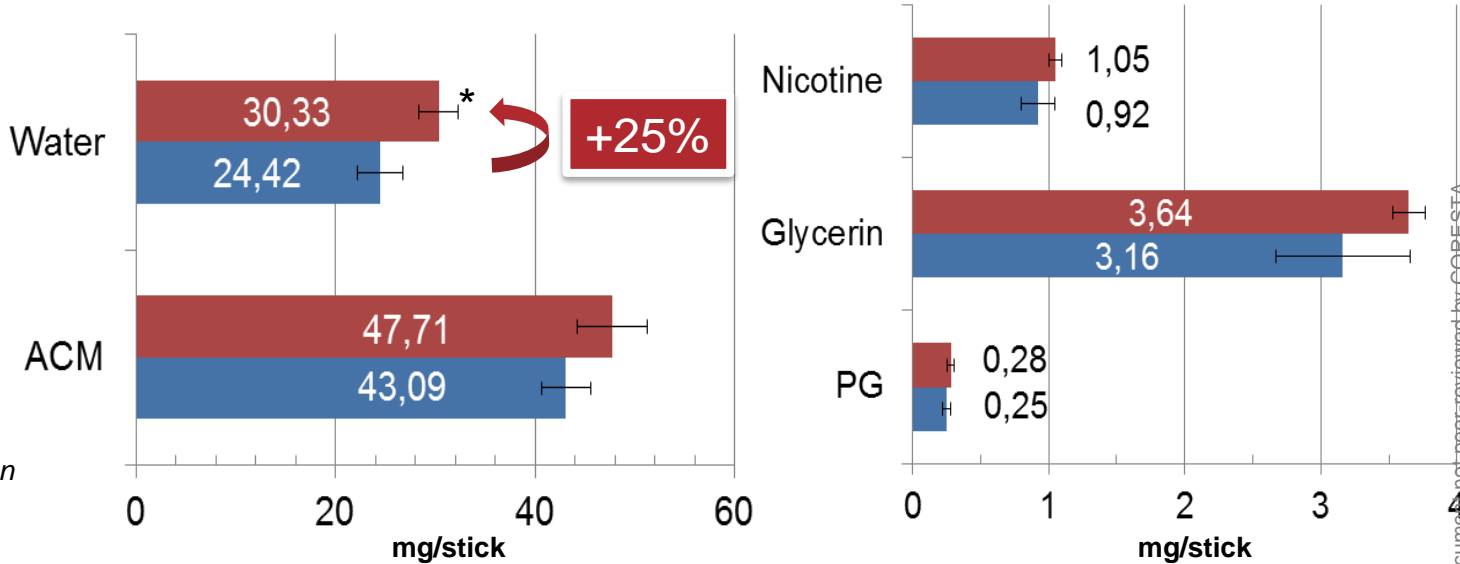


4. Effect of stick conditioning : results

Aerosol composition in mg/stick : mean \pm CI_{95%}
HCl smoking with 15s puff interval, 12 puffs, vents unblocked, n=4 replicates

■ 48h ISO3402
■ Sealed pack

HEETS Amber
in IQOS®



Significant increase of water in ACM when conditioning sticks in ISO3402 conditions

4. Effect of stick conditioning : results

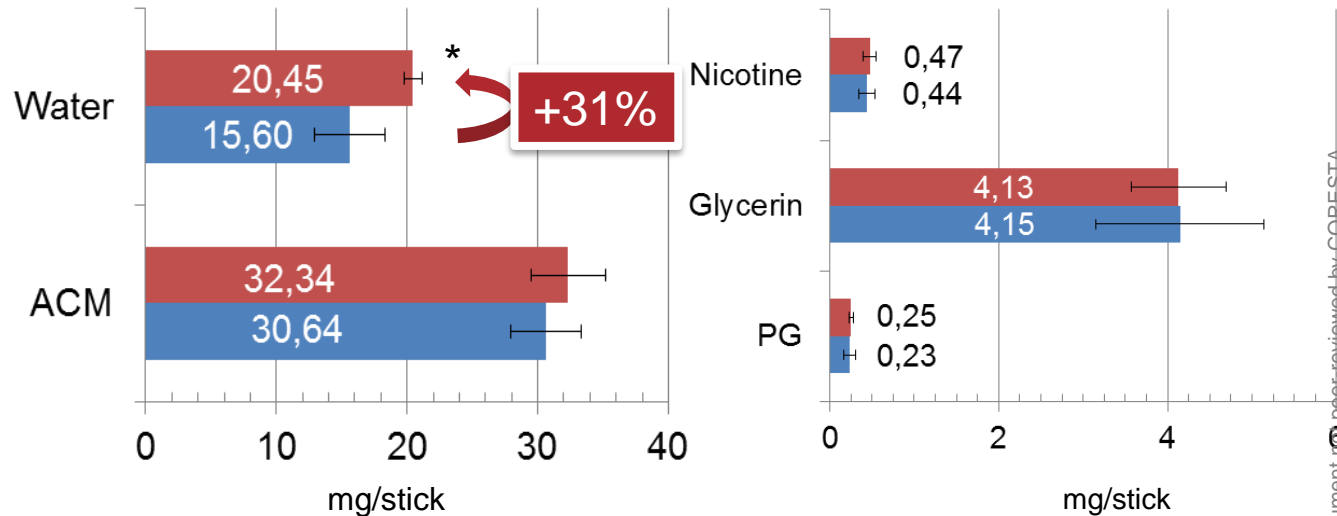
Aerosol composition in mg/stick : mean \pm CI_{95%}
HCl smoking with 15s puff interval, 12 puffs, vents unblocked, n=4 replicates

■ 48h ISO3402
■ Sealed pack

KENT neostiks™
glo™ G002



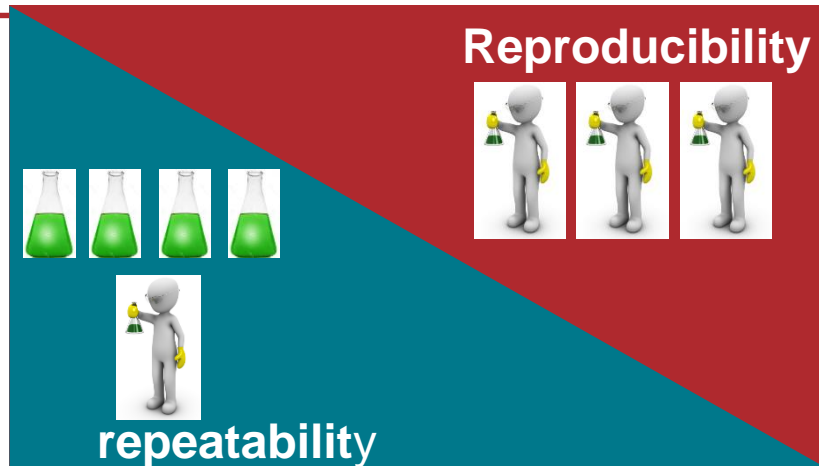
* statistically significant variation



Significant increase of water in ACM when conditioning sticks in ISO3402 conditions

5. r & R study : definition

N = 5 operators
4 days of analysis per operator
(4 replicates per day/1 mean value per day)



Sampling : Heets Amber sticks (France) in IQOS[®] G2 :
- sticks from sealed pack
- 3 sticks heated per replicate

HCl modified smoking regime : puff interval duration 15 s
12 puffs per stick (no cleaning puff)

6. r & R study : results

(mg/stick)	PG	Glycerin	Nicotine	Water	ACM
mean	0,27	3,91	1,08	22,37	40,67
IC _{95%}	0,04	0,53	0,14	5,07	3,12
Re %	13,60	13,50	12,90	22,70	7,70
Sr	0,017	0,212	0,060	2,152	1,457
SR	0,017	0,246	0,065	2,379	1,463
r	0,050	0,590	0,170	6,030	4,080
R	0,050	0,690	0,180	6,660	4,100

Relative error below 15%*, except for water
Higher contribution of repetability vs reproducibility

* According to AAPS journal, *Stability Recommendation for Best Practices and harmonization from the global bioanalysis consortium harmonization team* vol 26 n°3, May 2015

6. Conclusions

Effect on aerosol composition

✓ Puff interval : 15 s vs 30 s (HCl smoking regime)

No significant variation for total ACM, nicotine, humectants and water

✓ Tobacco stick conditioning ISO 3402 vs sealed pack (HCl smoking regime 15 s puff interval)

No significant variation for total nicotine, humectants, ACM

Significant water increase

6. Conclusions

HTP smoking method

- ✓ Method relative error <15% for ACM, nicotine & humectant except water
- ✓ Repeatability improvement : increase replicates number to 6.
- ✓ Water accuracy improvement : need to work on collection & extraction

Thank you

- SWM LTR lab for smoking analyses
- Diane Raverdy-Lambert, Stéphane Rouillard
- Philippe Le Men and Olivier Brenner for the statistical support
- Franck Binard for the temperature profiles