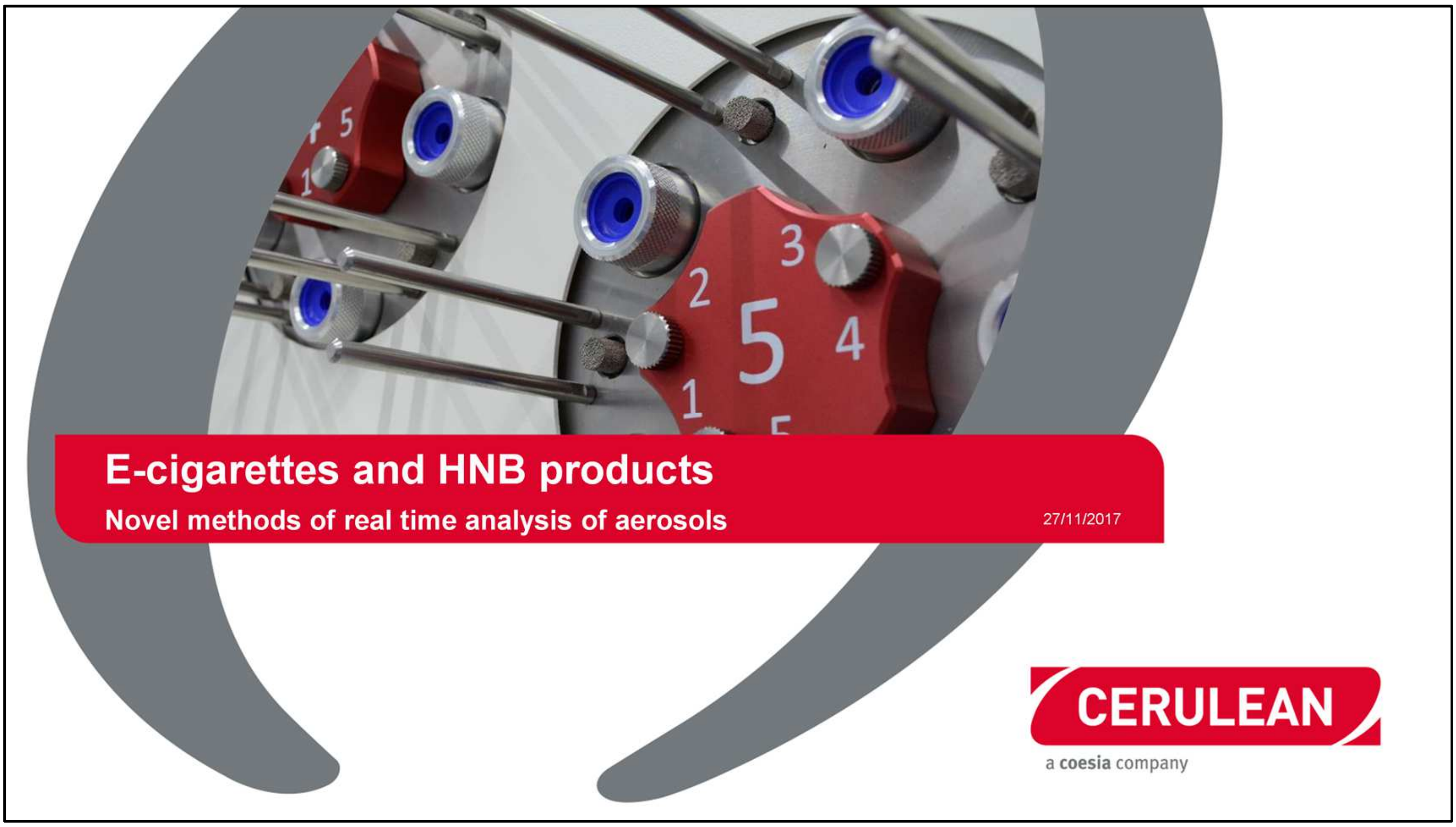
This presentation was delivered to an audience at the 12th International Tobacco Testing Technology Seminar held in Zhengzhou by Dr Ian Tindall (Head of Innovation and Marketing, Cerulean) and Mark Liu (General Manager, Cerulean China).



This presentation is on the subject of novel methods of real time analysis of aerosols from e-cigarettes and heat not burn products. We recognise that in China these products have not yet gripped the imagination, or the market share, that they have in the USA, Japan or Europe but perhaps this will be of some interest.

In particular we wish to focus on the use of Fourier transform infra red spectroscopy as a real time tool for the analysis of heat not burn and vaping products.

Introduction

Nicotine delivery systems are undergoing a revolution:

- Vaping devices
- Heat not burn sticks
- New regulatory frameworks (USA, EU)
- Campaigns for “smoke and tobacco free world” (WHO)

Do new nicotine delivery systems require new, more appropriate methods of analysis?

We would like to start by acknowledging my debt of thanks to researchers at Phillip Morris International who have reported on real time analysis and also to our co-workers at Cerulean, who have gathered the fundamental data behind the presentation today.

Nicotine delivery systems are undergoing a revolution. This has been driven by consumer demands for safer and more socially acceptable products along with the need to closely regulate the tobacco market.

This new environment allows us to look at more than just products. A revolution in product design could be accompanied by a revolution in how we view the analysis of emissions.

Vaping vs Burn Down

Traditional tobacco	Vape products
Yields per stick – delivery varies per puff	Uniformity of delivery through many puffs
Small set numbers of puffs to end point	Many puffs, subjectivity of end point
Principle components: Nicotine, CO and “Tar”	Principle components water, Nicotine, VG, PG
Secondary components – 4000+	Secondary components – flavours? Breakdown products when stressed?
Dirty process – ash, tar	Clean process
Very dependant on environment in which smoking takes place	?
Captured TPM and volatile component sample handling critical	?

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This table is one possible assessment of the differences between conventional burn down smoking and vaping. It is designed to support the view that vaping is so different from conventional smoking that we can radically change the way we analyse the aerosol from the device.

We have left two question marks on the vaping side of the table because we think greater understanding is needed before a definite conclusion can be reached, but there is some evidence to suggest that sample handling and environmental conditions are less critical in vaping than in conventional smoking.

We should note that heat not burn products lie somewhere between the two although in many respects they tend toward the vaping model.

Other comparisons might deal with the relative water content of conventional cigarettes and these new generation products, or the formation of carbon monoxide during combustion or the process of pyrolysis. The list of differences may not be endless but it is certainly a long one.

FTIR – Fourier Transform Infra Red Spectroscopy

What it offers

- Real time quantitative analysis of multiple aerosol components
- Large array of potential analytes

Disadvantages

- LOD and LOQ generally higher than for alternative methods
- Needs direct coupling to a device under test
- Calibration requires a library of materials that have been characterised
  - OK for main components but less so for flavours

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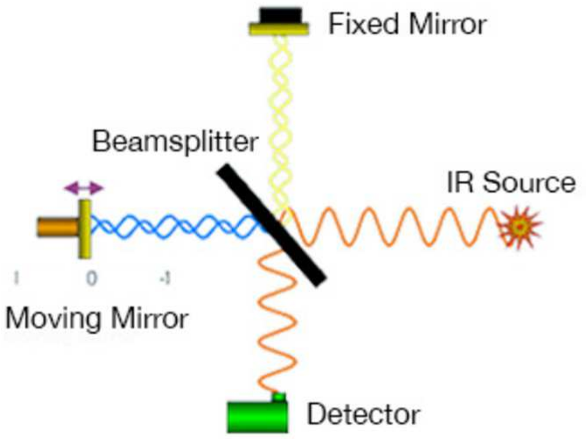
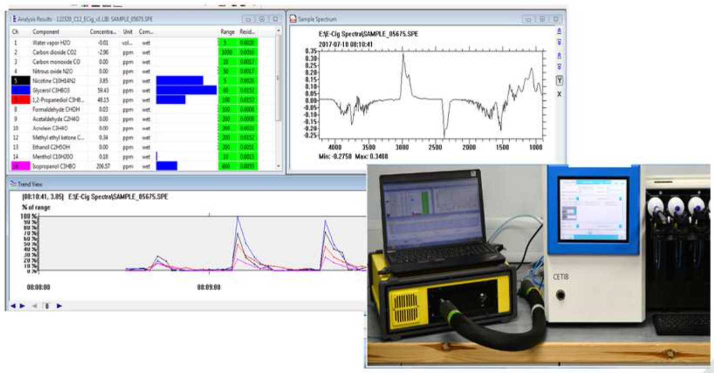
We want to talk to you about the coupling of a Fourier transform infra red spectrometer (an FTIR) directly to a vaping machine so that there is direct analysis of the aerosol formed, and the power this gives us at a technique. This is not a commercial product as yet - we am not selling anything here – but it is currently in the laboratory assessment phase and currently showing some promising initial results. The use of such an analyser is possible because of the relative cleanliness of aerosol generated from these new nicotine delivery products. This allows us to draw the aerosol directly into the puff engine and then exhaust it into the analyser in a chemically and physically unchanged form. The real advantage of this technique is that for major components of these aerosols such as nicotine, water, propylene glycol and vegetable glycerine you can get quantitative data almost instantly on a puff by puff basis, a requirement that is implied by the tobacco products directive of the EU which demands uniform delivery through the life of a product. Moreover the technique can give real time puff by puff quantitation of a number of key chemicals of interest that are usually difficult to capture and handle such as formaldehyde and other carbonyls.

There are disadvantages to this technique such as the LOQ and LOD being generally higher than for more convectional techniques and the coupling to a vaping machine is not quite straightforward. Most critically a library needs to be built up of the response of the analyser to different chemicals in the gas or vapour phase as this forms the basis for subsequent analysis. We would like to show you three simple experiments that show the versatility of this technique, in all cases the results of the tests were obtained in real time. The analysis of the data took a little longer.

What is FTIR?





We should start by gaining an understanding of Fourier transform infra red analysis. Do not expect too detailed an explanation, that would be the subject of a whole university course.

FTIR offers quantitative and qualitative analysis for organic and inorganic samples. Fourier Transform Infrared Spectroscopy identifies chemical bonds in a molecule by producing an infrared absorption spectrum. The spectra produce a profile of the sample, a distinctive molecular fingerprint that can be used to screen and scan samples for many different components simultaneously.

In FTIR spectroscopy, a beam of light is passed through a series of mirrors that cause the beam's individual wavelengths to hit each other in a way that allows a sample to absorb some wavelengths, while others are blocked. Light absorption is measured and a computer infers the absorption rate of each wavelength within the beam.

FTIR spectroscopy measures the absorption rate of many different frequencies simultaneously and through use of libraries can deconvolute complex mixtures for qualitative and quantitative analysis.

Intensive Vaping – Changing Puff Parameters

- FDA requires “intensive vaping” without specifying what this means
- Current CORESTA thinking is that this regime is defined by the tester depending on product under test with a justification for the parameter chosen
  - We can use experience to decide on an intense regime but how scientifically justifiable is our judgement?
  - If we have to provide evidence for our decision how rigorous must we be in devising a test and how much time and cost will this incur
  - If we change vaping parameters from the ISO 55ml, 3 second, 30 second interval square wave does this change nicotine delivery?
- In line FTIR provides a quick method of determining nicotine delivery as we change vaping parameters of duration, volume and interval

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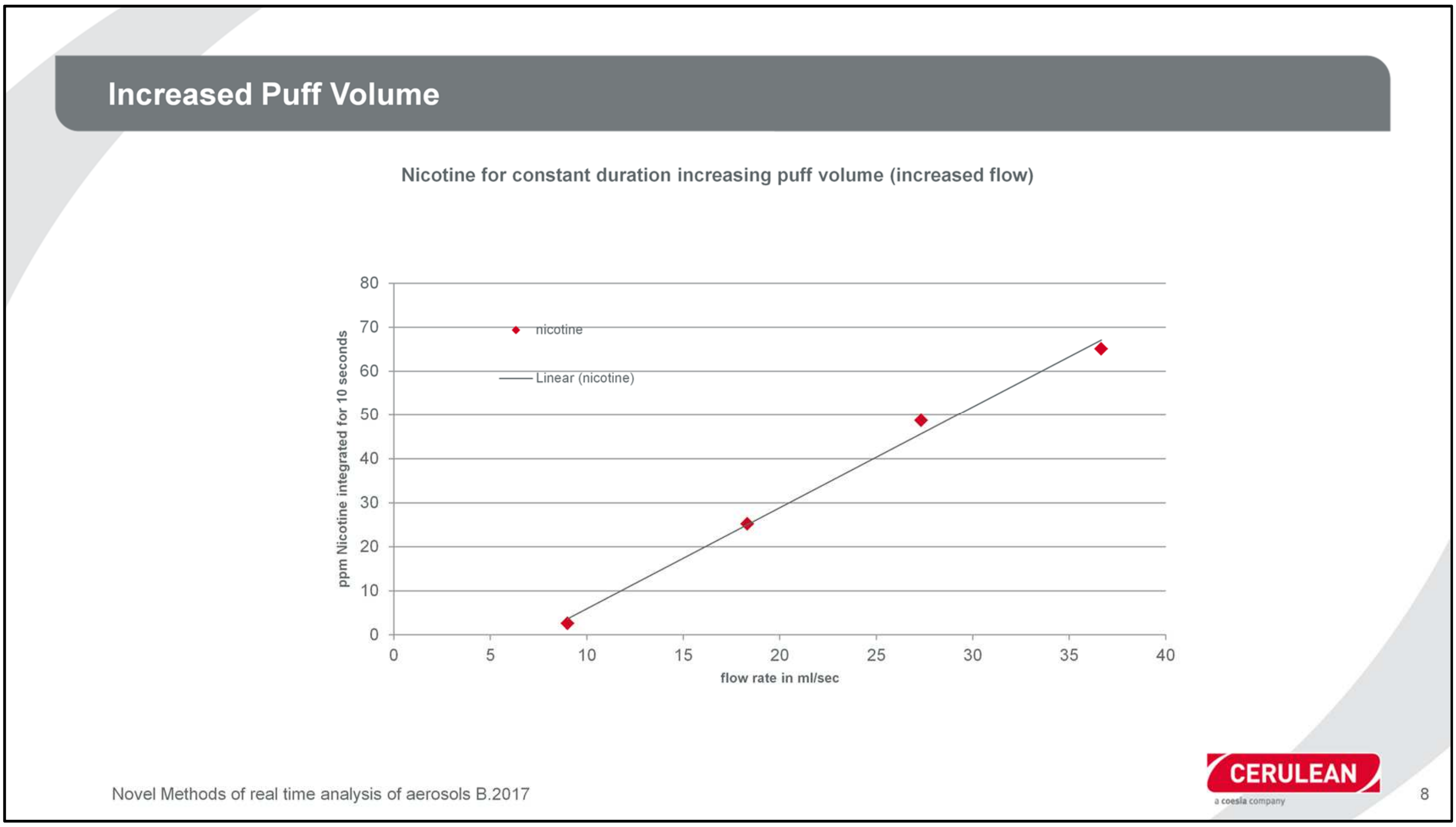
There are many challenges we face with novel nicotine delivery devices as they are introduced to the market. There are legislative challenges and challenges around the development of analytical methods that support both the product development and product stewardship agendas.

Today the American FDA is setting the pace when it comes to legislation. Within the so called deeming regulations there are requirements for intensive vaping without defining what this means.

The CORESTA EVAP sub group has been considering this with the possibility that any recommendations made by the group may in time become the basis of an ISO standard. However their main conclusion is that although human topography studies may give guidance as to what is an intense regime this will be device specific.

This has led to the situation where individual scientists will define their intense vaping protocol and have to be able to justify the decision they made on the basis of reason and science.

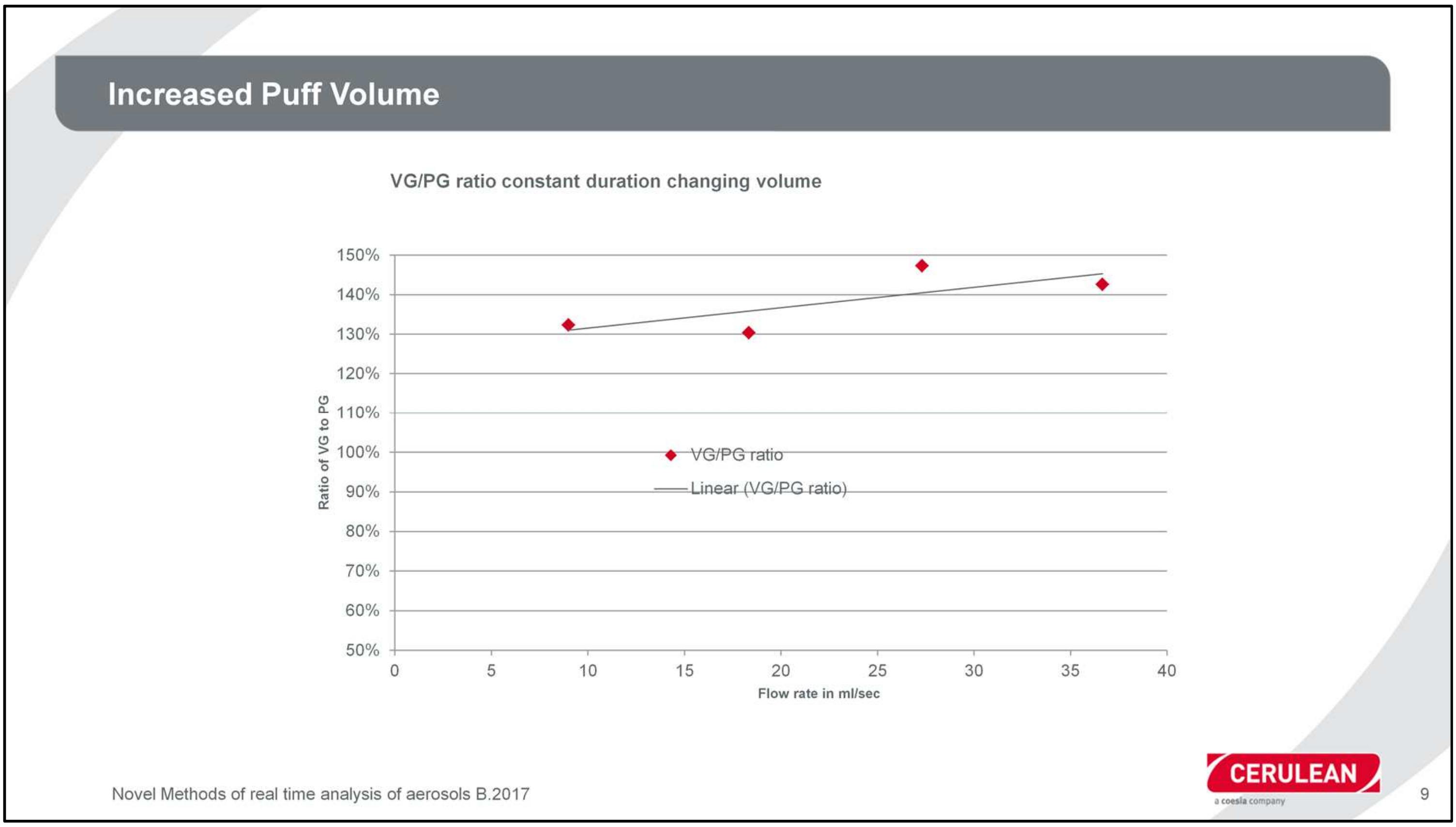
The problem remains as to how valid is the judgement of an individual? In this, rapid testing with an FTIR system may give credence to the decision made concerning the selection of an intensive regime.



For this experiment we used a Cerulean CETI8 vaping machine and a Gasmet FTIR analyser supplied by a company called Quantitech. The exhaust from the puff engine was connected via a heated transfer tube to the FTIR and the e-cigarettes were vaped without a CFH in place. Some of our earlier work showed that the transfer efficiency of this arrangement was better than 93% and than the aerosol particle size was well distributed about the 0.3micron size and was unchanged by the transfer through the system. This was a key starting point for using this analyser as we had to make sure that we did not introduce experimental artefacts that would render our findings useless.

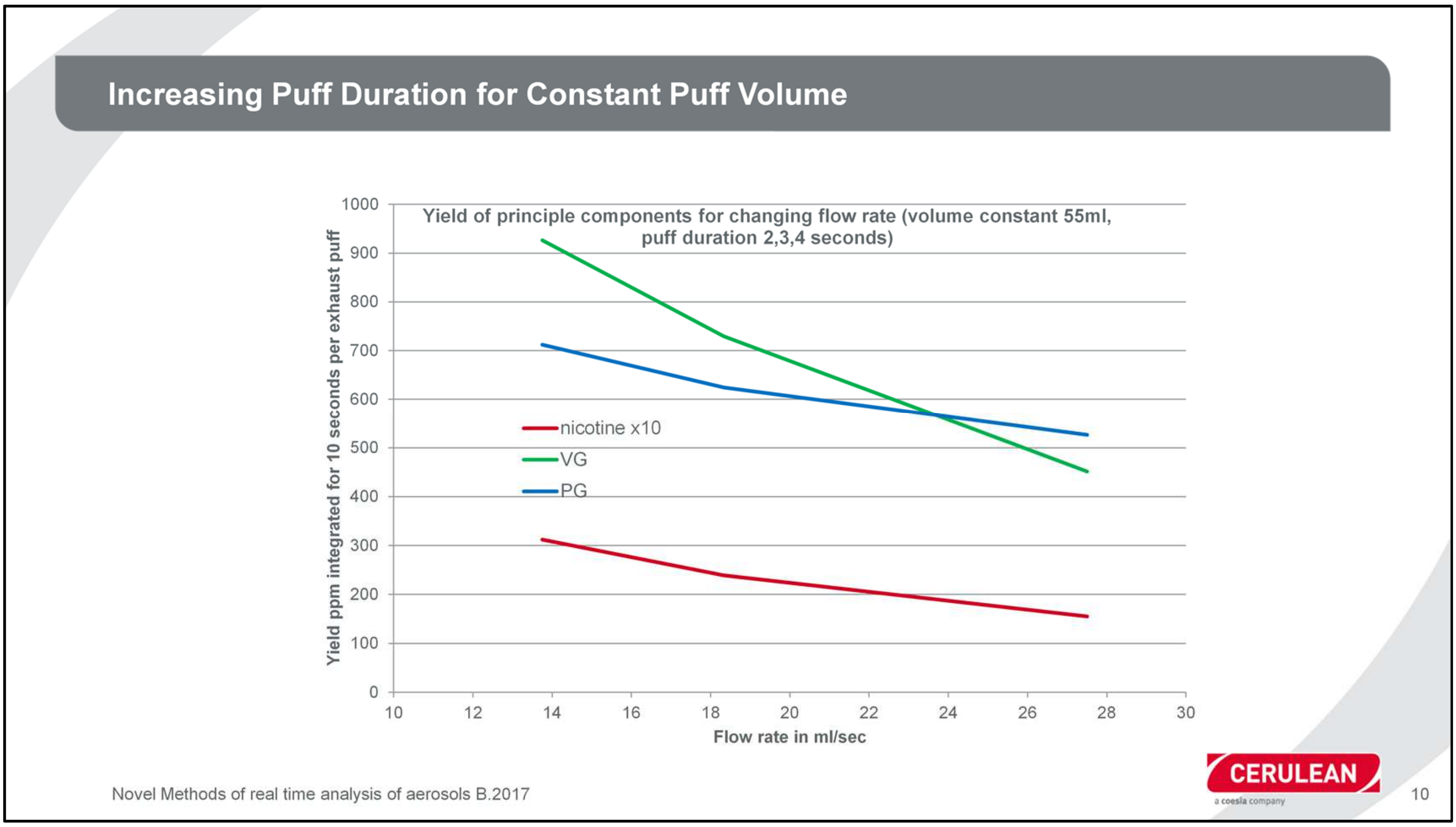
The data shown here was gathered in a single 4 hour period, each point representing 10 puffs from 4 devices. The major part of the experimental time being taken in adjusting the temperature of the transfer tube to an optimum.

Simply increasing the puff volume, in effect increasing the flow rate, gave a simple relationship between nicotine delivery and flow rate. As we know from the health Canada smoking protocol this is not necessarily the case for all traditional cigarettes.



Slightly less expected was the change in the propylene glycol to vegetable glycerine ratio with increase flow rate or puff volume, at end of life this could have a impact on the production of minor components of the vapour matrix that are formed as a result of the decomposition of PG or VG.

Again there is a simple ratio of change that could be used for predictive purposes.



Here it can be seen that the effect of increasing puff duration, or the reduction in flow rate on the yield of the principle components of the vaping aerosol. This is not quite such a clear linear relationship although the necessarily small number of points may be distorting the graphs.

It would, in theory, be possible to produce a set of conditions where all parameters are varied with appropriate reference to topography studies and so give a maximum yield. This could form part of a designed experiment and support any decision made on the selection of vaping parameters for intensive vaping.

Better still it would be possible to predict the yield of the Nicotine, PG, and VG delivery for alternate conditions on the device based on a standard ISO experiment and so not needing to actually vape the product under these conditions thus saving laboratory analysis time.

This would be applicable for other more traditional analysis methods but the beauty of the FTIR method used here is that we can validate our assumptions quickly and perform a cross check confirmation measurement in a very short space of time.

Delivery Uniformity

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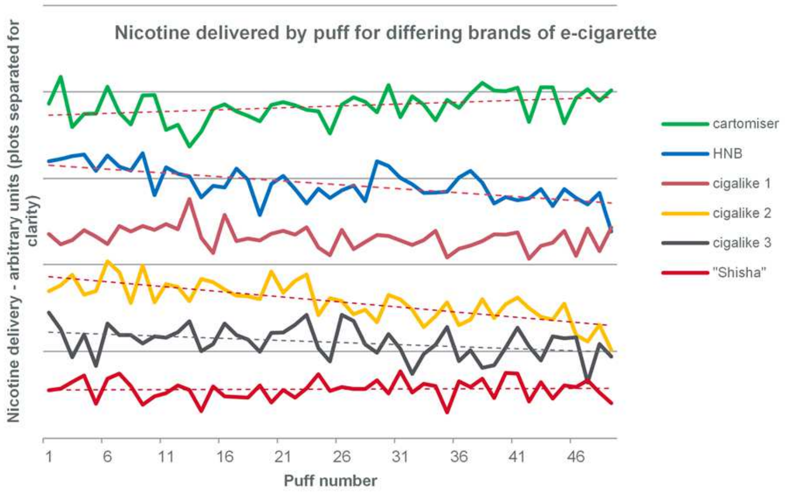
- Nicotine should be delivered uniformly throughout product life
- Puff-by-puff is the best way to show this
- Different brands and devices show different behaviours
- HNB and vaping products benefit from the FTIR system



A more routine application could be the assurance that nicotine is being delivered uniformly throughout a product life as a demanded by EU regulations.

This is something that varies from device to device and also from batch to batch of what is nominally the same device and so this becomes both a QA requirement and a product development requirement. The product developer wants a product that has the capability to consistently deliver nicotine and the quality team want to be sure that what has been produced meets the design specifications in consistently delivering nicotine in the vapour stream.

Puff by puff determination is simple with this system and shows in this example the different behaviours of different devices.

This again was carried out on an 8 port vaping machine and the experiment was conducted over 2 hours with the data being generated in real time. As we can see the different devices varied in the consistency of nicotine delivered, one disposable e-cigarette delivered less nicotine through its life, the others delivering fairly consistent yields of nicotine. In contrast the cartomiser delivered more nicotine as it was used. The heat not burn device used in this experiment dropped in nicotine delivery which was probably to be expected as these essentially distil nicotine from tobacco.

The most uniform delivery was from one of the cigalikes and from something described as an e-shisha pen.

Delivery Uniformity

Product	Nicotine			Glycerol			Propylene Glycol		
	Mean	SD	COV	Mean	SD	COV	Mean	SD	COV
Cartomiser	9.2	0.882	0.096	228.5	12.797	0.056	155.4	6.251	0.040
HNB	4.7	0.989	0.211	139.4	31.00	0.222	846.8	92.920	0.110
Cigalike 1	2.6	0.642	0.250	231.9	47.688	0.206	228.7	30.822	0.135
Cigalike 2	2.9	1.116	0.384	276.6	92.925	0.336	173.8	52.953	0.305
Cigalike 3	5.5	0.913	0.165	197.1	29.528	0.150	400.5	37.554	0.094
E-Shisha	1.8	0.569	0.312	235.1	15.116	0.064	311.6	12.660	0.041

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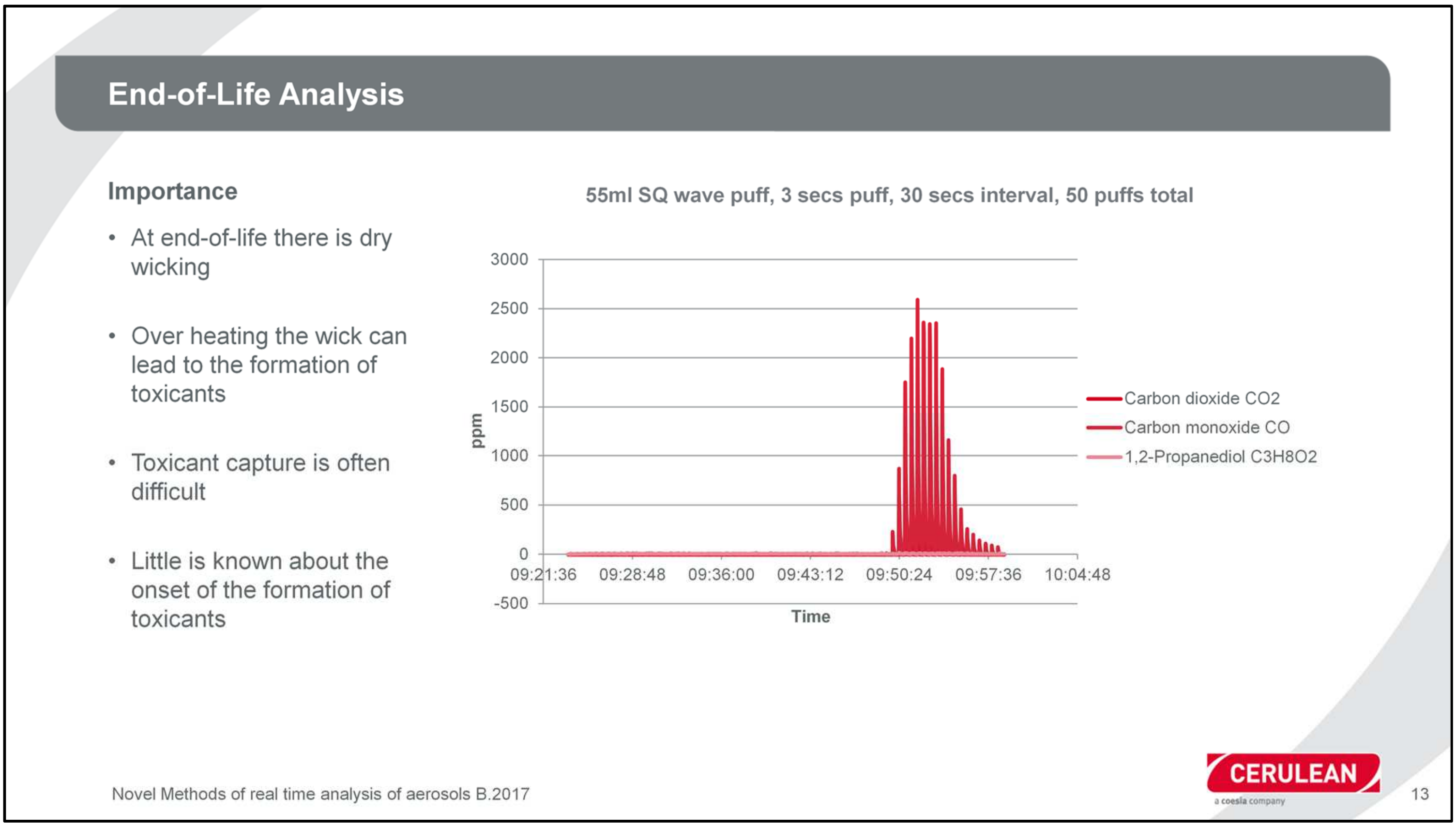
But as we can get quantification of multiple analytes simultaneously using FTIR, and the technique is quantitative we can not only track the nicotine delivery on a puff by puff basis but also look at a range of compounds of interest and get true quantitative delivery information during the same experiment.

The table here shows the three main components of vaping products, this is the same experiment as shown on the graph and the data was collected at the same time. Please note we not only got propylene glycol and glycerine from the conventional e-cigarette products but we also get significant propylene glycol and glycerine from the heat not burn product tested.

The COV for these compounds for all products is acceptable for diagnostic purposes, units are parts per million which can be related routinely to weight of delivery with some simple mathematics.

With this quantitative information we can reject the hypothesis that the constancy of delivery of nicotine through the puff life is related to the concentrations of the other major components of the vapour stream such as glycerine and propylene-glycol.



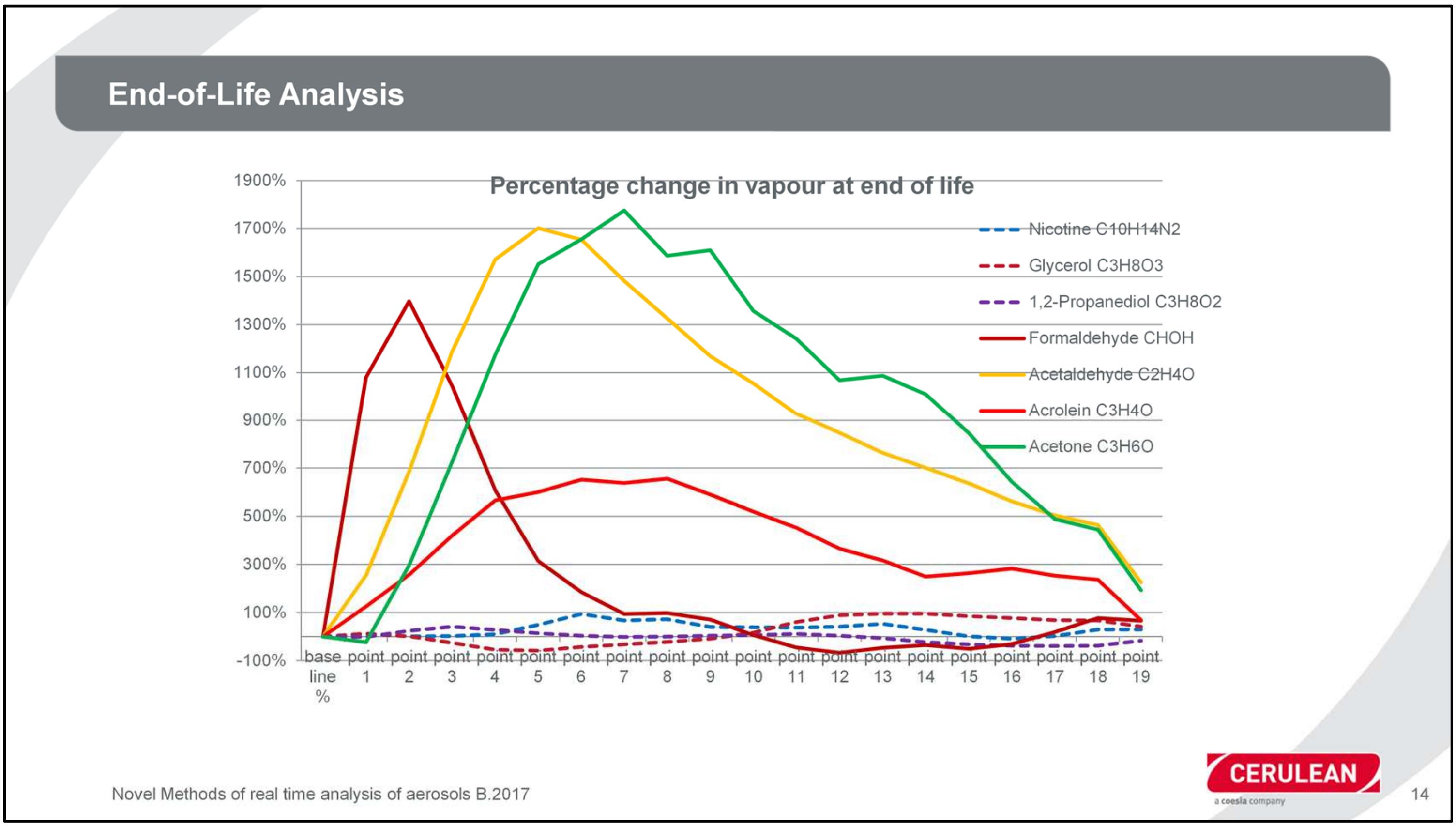
It is known that at the end of life of vaping products the composition of the vapour changes as the wick dries and becomes directly heated. This has been the subject of a number of scientific papers.

This is known as dry puffing and it produces an acrid taste and has the potential to expose the user to thermal degradation products. Some of these breakdown products can be injurious to health and an understanding how these build up on a puff by puff basis can lead to improved vaping product designs.

A gross marker of this onset of dry wicking is the formation of Carbon Monoxide, essentially a combustion product. Now we do not need a FTIR to determine if CO is being made, there are many other analysers that would be just as effective and a lot cheaper to couple to a vaping machine for puff by puff analysis, but the FTIR analyser can quantify CO and also other thermal degradation products directly.

For the graph shown here a tank type e-cigarette was filled with commercial e-liquid and allowed to soak the wick. The surplus e-liquid was drained from the tank and then the device set to vape on a CETI8 vaping machine.

The x-axis is a time profile rather than a puff count but we can see there is a very sharp transition between forming little or no carbon monoxide and substantial amounts of CO being formed, please note the occupational exposure limit for CO in the UK is 200 ppm on a time weighted average basis with an absolute instantaneous exposure limit of 1500ppm. This is exceeded here by over 30%.



As has been noted e-cigarettes can produce toxins such as formaldehyde although this has been shown to be principally linked with dry puffing conditions. With the FTIR analyser we are capable of looking at the formation of carbonyls as dry wicking occurs. Again the experiment was conducted using a specially prepared tank system and the composition of the aerosol monitored on a puff by puff basis. The experimental protocol including providing data for study and took approximately 2 hours requiring no special capture or treatment of the aerosol as this was passed directly into the FTIR from the CETI8 puff engine.

Looking across the wider range of chemicals analysed we can see a marked change in the chemical composition of the vapour as we begin to dry wick. Here three points are taken together as a moving average to smooth the peak data as we are concerned with trace concentrations in some instances. I have also shown this as a percentage change from normal baseline conditions to enhance the changes and place all data on a single set of axis. It can be seen quite clearly that carbonyls such as formaldehyde, acetaldehyde and acetone increase significantly as the wick dries and puffing continues. Nicotine delivery also increases during this end of life phase as VG decreases. A full life experiment could be conducted where a filled cartomiser or disposable e-cigarette is puffed to and beyond exhaustion and so determine the maximum puff count that can be delivered before the formation of toxins occurs. With the sophistication of these devices it may be possible to make this an inbuilt limit on the device and so make the product safer in use. Further chemical species could be examined such as the tobacco specific nitrosamines but this would require the production of library spectra, something we did not have time to explore in these experiments.

Concluding Summary

- New product types deliver nicotine in a way that differs from conventional burn down products
- Some of the technical restrictions on using different analysis methods are removed with these new devices
- One possibility is the real time analysis of multiple elements of the vapour aerosol
- This can be achieved with Fourier Transform Infra Red analysis coupled directly to a CETI8 Vaping machine
- This approach gives a rich set of quantitative and qualitative data in a fraction of the time taken for similar analysis using “cigarette methods”
- The data can be used effectively for QA, product development or competitor analysis purposes

Thank you for your attention

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