Research Study Publication

SUCRALOSE
Toxicological issues related to its use in vaping products

by OpenSciences.

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SINCE 2014,

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1. INTRODUCTION

For some years now, vaping products with sweet flavours have been increasingly popular with vapers and more particularly with young users[1].

In response to this high demand, some liquid manufacturers have chosen to use sucralose (CAS 56038-13-2) in the composition of their products, because of the physical-chemical properties and the intensity of the sweet taste of this synthetic sweetener.

Sucralose (E955) is mainly used as a sugar substitute in agro-food products[2]. This chlorinated organic compound obtained by the selective chlorination of sucrose (table sugar) (*Figure 1*) has a sweetening power between 400 and 700 times more intense than the latter[3,4].

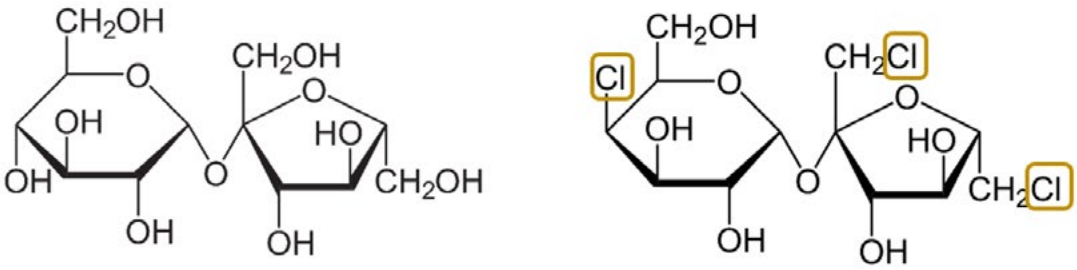

Figure 1 - Chemical representations of the structure of sucralose (right) and sucrose or saccharose (left).

The French standard with voluntary application XP-D90-300 part-2[5], on the composition and manufacturing requirements of vaping liquids, has a list of sweeteners whose use is prohibited or limited. Currently, sucralose is not included in this list.

However, the new consumption method of the molecule proposed by the popularity of sweet vaping products raises new issues for its agro-food use.
Is sucralose stable in the heating conditions provided by a vaping device? What are the toxicological consequences of its presence in a liquid for vapers? Are the benefits of its use in vaping products greater than the risks that it creates?

Moreover, the Ingésciences laboratory wanted to carry out a study to attempt to answer these questions. The first part of the study proposes to explore, through a bibliographic summary, the toxicological aspects related to the consumption of sucralose and more particularly those associated with its use in vaping. In the second part, the data of the literature will be compared with the experimental study carried out in the laboratory intended to observe the behaviour of sucralose during its vaporisation using different personal vaporisers.

2. BIBLIOGRAPHIC SUMMARY

a. Intrinsic toxicity of sucralose

Traditionally used in agro-food products, the gastro-intestinal ingestion and absorption of sucralose have been the most studied. At the start of the 2000s, the sweetener is mainly considered as non-toxic in ingestion[6]. Thus, in 1999, the American Food and Drug Administration (FDA) approves its use in agro-food products and fixes a maximum daily dose consumed at 5 mg/kg of body weight[7]. In Europe, from 2004, a directive authorises the use of this sweetener in agro-food products and fixes a permitted maximum daily dose of 15 mg/kg[8]. Whether in the United States or in Europe, no consumption restriction is applied especially concerning pregnant or nursing women, diabetics or people receiving medical treatment.

With the popularity of its use, recent studies are questioning the compound's safety. Indeed, sucralose intervenes in and influences many physiological functions once absorbed by the organism[2].

Thus, it appears that sucralose consumption changes glycaemic regulation and certain physiological mechanisms involved in body weight regulation[9,10].

It also affects the assimilation and metabolization of certain pharmaceutical active ingredients pointing to the reduced effectiveness of certain medical treatments[11].

The metabolic pathways specific to sucralose have been relatively well established but the effects of its metabolization products with regular consumption remain unknown[12].

More worryingly, there are doubts about the possible mutagenic nature of sucralose. It would be capable of altering the structure and integrity of DNA in animals[13].

This non-exhaustive list of the physiological effects caused by sucralose consumption contrasts then with the current regulation covering the use of this molecule in agro-food.

However, the research carried out on sucralose often deals with gastro-intestinal consumption since this compound is used in products intended for ingestion. To date, little data is available as to the effects in inhalation of sucralose.

Certain studies published in the scientific literature are nevertheless focussed on the thermal stability of sucralose because of its presence in certain heated agro-food products.

b. Thermal stability and toxicity of the degradation products of sucralose

Not long after the approval of the use of sucralose in agro-feed products, much work was committed to studying its thermal stability. First appearing as safe, its presence in current heated consumer products has rapidly revealed the low tolerance of the molecule to temperature[14].

Indeed, found in the literature are studies dealing with the thermal stability of the chemical structure of sucralose in fairly harsh heating conditions (>250°C)[15]. In these extreme conditions, the molecule hydrolyses (breaks up), and produces chlorinated toxic compounds especially members of the chloropropanol family.

In 2015, a research team also demonstrated, from several analytical techniques, the thermal instability of sucralose[20]. Subject to a heating temperature greater than 120°C, i.e. a temperature less than its melting point (=130°C), sucralose produces chlorinated compounds like hydrochloric acid, chloroacetaldehyde, polychlorinated aromatic hydrocarbons and molecules of the chloropropanol family.

Certain molecules of this chemical family (chloropropanols), more particularly 3-chloro-1,2-propanediol (3-MCPD; CAS 96-24-2), have high toxicity and a carcinogenic aspect for humans[16].
From 1999, Hutchinson et al.[17] show that sucralose in solution at different pH (acid, neutral, basic) and heated for 1 hour is fully degraded at a temperature of 180°C. Different degradation products such as furans and their derivatives are observed as well as acidification of the medium. More specifically, the release of hydrochloric acid starts at 119°C and participates in the formation of 3-MCPD[18].

The formation of chloropropanols from sucralose in the presence of glycerol was shown in 2010[19]. Indeed, Rahn et al. detected the presence of 3-MCPD and 1,2- and 1,3-dichloropropanols (1,2-DCP & 1,3-DCP) during the pyrolysis of sucralose in the presence of glycerol.

All these items demonstrate the thermal instability of sucralose accompanying the production of toxic species. It also drives certain scientists to strongly discourage use of the molecule in any process involving a temperature greater than 120°C[20].

This bibliographic data also indicates that the degradation phenomena of sucralose could arise with its use through a vaping device. Since personal vaporisers require e-liquid heating temperatures between 189°C and 292°C[21], it appears essential to study the behaviour of this molecule in heating conditions representative of those inherent to its use.

c. Toxicological aspect of sucralose in vaping

Because of the increasingly frequent use of sucralose in vaping products, the scientific community is starting logically to study its role and the toxicological consequences of its presence in products linked to smoking cessation. This preliminary work points to the risks run by the presence of sucralose in vaping liquids.

A publication dealing with the «sucralose/e-cigarettes couple» was listed in 2017[22]. Rosbrook et *al.* had the aim of determining the quantity of sucralose transmitted to the vaper and whether this was sufficient to detect the sweetened flavour sought by vapers.

They showed that the type of equipment used had an influence on the delivery of sucralose to the vaper. Indeed, in the vaporisation conditions selected by the authors, transmission of the sucralose contained in the liquid to the aerosol generated by its vaporisation appeared ineffective: only 10% of the quantity of sucralose of the liquid was found in the vapour, assuming low volatility for the molecule.

This observation can be explained in various ways:

- Sucralose vaporises very poorly in the study conditions and remains in the heating element or in the reservoir of the vaporisation system.
- Sucralose is not thermally stable and degrades during the heating required to vaporise the liquid.
- The equipment selected for the study favours one or the other of the two hypotheses above.

However, the study does not show results related to a chemical measurement of the emissions or a possible search for degradation products specific to sucralose.

A second more recent publication (May 2019) reports on an analysis of the emissions produced from sucralose products[23]. The authors propose a study more suited to recent vaping products, conveyed by a choice of vaporisation conditions representative of the current market (clearomizer, Subtank Nano from KangerTech fitted with a resistance of 1.2 ohm with vaporisation power 20 watts).

After a measurement of the average concentration of sucralose present in market products (between 2.7 & 6.4 mg/mL), the authors choose to compare the emissions of custom liquids to contain a scale from 0 to 5.4 mg/mL of sucralose.

This work allowed them to produce different observations:

1) The quantities of certain specific degradation products of the propylene glycol /vegetable glycerine (PG/VG) matrix of the liquid (propanol CAS: 123-38-6, acetaldehyde CAS: 75-07-0, acrolein CAS: 107-02-8, formaldehyde CAS: 50-00-0 and glycolaldehyde CAS: 141-46-8) are greater after vaporisation of a solution containing sucralose.

2) The vaporisation of sucralose using a personal vaporiser is accompanied by a release of acids as some previous publications had suggested[19,20]. The acids produced (hydrochloric acid, acetic acid, etc.) change the form of the nicotine present in the aerosol. Indeed, the presence of acids in the aerosol transforms 25% of the nicotine under its ionised form after vaporisation.

3) The production of certain specific degradation compounds of sucralose is also observed. 3-MCPD, result of the reaction between hydrochloric acid and VG, is identified (*Figure 2*) like all other chlorinated organic compounds such as 1,6-dideoxy-1,6-dichlorofructose (CAS 69414-08-0).

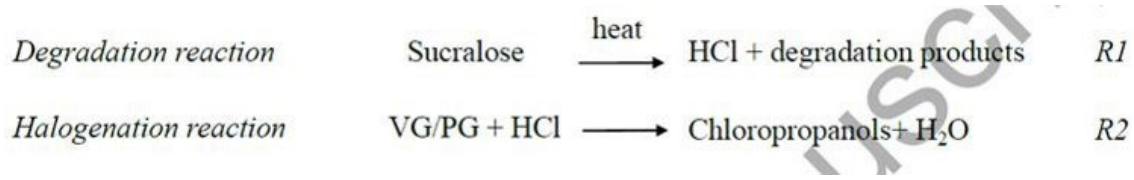

Figure 2 – Chemical reactions acting during the heating of sucralose in the presence of glycols[24].

Degradation of the cotton present in the heating element of the vaping device is also favoured by the presence of sucralose. This is indicated by an increased presence of levoglucosan (CAS 498-07-7), a specific degradation product of the pyrolysis of cellulose, in the emissions of sucralose liquids. Catalysed by the acid conditions, degradation of the wick can then be indirectly related to the release of acids coming from the heating of the sucralose. This phenomenon explains the decreased lifetime of the heating elements observed during the consumption of this type of product.

4) Finally, the greater the concentration of sucralose in the liquid, the greater the quantity of all the mentioned degradation products.

A third publication published in July 2019 attempts to study quantitatively the production of two chlorinated compounds, 3-MCPD and 1,3-dichloropropanol (1,3-DCP; CAS 96-23-1) during the vaporisation of a sucralose liquid[24].

Initiated and in part funded by the «Food and Drug Administration» (FDA), the study shows the formation of 3-MCPD and 1,3-DCP in soft vaporisation conditions (2.2 ohms; 5 & 11 watts) as well as in extreme conditions (0.15 ohm; 100, 150 & 200 watts). Surprisingly, the concentration of sucralose present in the liquid influences the quantity of degradation products much more than the conditions of vaporisation. In other words, the more the sucralose is concentrated, the more there is 3-MCPD and 1,3-DCP in the vapour, without considering the conditions of vaporisation.

While there is only little information on the toxicity induced by the consumption of 3-MCPD and 1,3-DCP[25], the IARC (International Agency for Research on Cancer) classes them as probable carcinogens for humans (group 2B)[26]. The toxicological risk of these two compounds is added to those induced by the «classic» degradation products found in the emissions of a personal vaporiser.

As far as we know, there is only one provisional maximum tolerable daily intake (PMTDI) covering the consumption of chloropropanols[24]. It is 2 µg/kg of body weight per day. According to the measurements of the authors and assuming consumption of 150 puffs per day, a vaper of 70 kg could be exposed to doses of chloropropanols between 0 and 18 times more than the PMTDI during the consumption of a sucralose product.

Faced with the recent conclusions of these scientific articles, it is more than reasonable to think that the consumption of a vape liquid containing sucralose, in any conditions, will entail a greater toxicological risk than the consumption of a liquid not containing it. While clear, the increase of this risk will be difficult to estimate and measure.

The above bibliographic items show that the use of sucralose in vaping products raises legitimate toxicological questions. Ingésciences has therefore carried out an experimental study in order to substantiate the results available in the literature.

3. STUDY OF THE VAPORISATION OF SUCRALOSE AND THE FORMATION OF 3-MCPD USING DIFFERENT VAPING DEVICES

The objectives of this experimental study are:

- To qualify the efficiency of vaporisation of sucralose using different vaping devices.
- To quantitatively evaluate the production of chloro-1,2-propanediol (3-MCPD) during the vaporisation of vaping products containing sucralose.
- To draw the main toxicological conclusions related to the use of sucralose in vaping products.

For this purpose, 3 vaping devices were used. For each of them, two chemical parameters are studied:

- Influence of the level of sucralose (1.5 to 10 mg/mL)
- Influence of the level of propylene glycol (PG) and vegetable glycerine (VG) in the liquid matrix.

a. Equipment and methods

i. Mode and parameters of vaporisation

During the study, aerosols were emitted from our robot vaper U-SAV. It makes it possible to simulate in the lab the behaviour of vapers, associated with the variability of equipment and the diversity of vape liquids and thus to study the composition of the aerosol in actual vaping conditions.

In this study, the robot vaper U-SAV ensures compliance with the vaporisation parameters of the standard AFNOR XP-D90-300-3[27] on the emissions of vaping products. The automaton allows us to obtain and to reproduce 5 series of 20 puffs of 3 seconds at a flow rate of 1.1 L/min. The series being each spaced by 300 s and puffs by 30 s.

The experiments were carried out using the different devices and resistances described in the table below:

Table 1: Summary of the devices and resistances used during the experiments.

Devices	Zenith (Innokin)	Cubis (Joyetech)	Veco Tank (Vaporesso)
Resistances (value of resistance/ material of resistance wire / type of wick)	0,5 ohm / Kanthal / Coton	1 ohm / Acier inoxydable / Coton	0,5 ohm / Acier inoxydable / Céramique
Power applied	12W	15W	30W

ii. Trapping method

The system used to trap the vapours is a cryogenic trap. Developed within our laboratory, it is capable of condensing the vapours generated by the robot vaper U-SAV at a temperature of -45°C.

In a single generation, this technique makes it possible to analyse and to look for the different compounds present in the aerosol. A specific rinsing protocol with methanol was used in order to optimise the extraction of the trapped molecules.

The efficiency of trapping the 3-MCPD by the cryogenic system was characterised by vaporising a liquid composed of PG/VG (50/50; v/v), nicotine (20 mg/mL) and 3-MCPD (5 mg/mL).

This was done for all the equipment chosen and described above.

The different average rates of recovery of 3-MCPD are given in *Table 2*.

Table 2: Summary of the average rates of recovery obtained for trapping the molecule 3-MCPD.

Equipment used	Zenith	Cubis	Vecotank
Average rate of recovery of the molecule 3-MCPD	82% +/.10%	89% +/.8%	83% +/.16%.

iii. The formulations studied

Throughout the operations, the liquids used were composed of 20 mg/mL nicotine. The nicotine analysed in the emissions acts as control and makes it possible to check proper running of the operation.

In order to study the influence of the level of PG and VG on the production of 3-MCPD, different liquids were made up ranging from a matrix 100% PG to 100% VG each containing 10 mg/mL of sucralose.

In order to evaluate the impact of the level of sucralose, several products were prepared in an interval of concentration ranging from 1.5 mg/mL to 10 mg/mL. For these liquids, the level of PG/VG 20/80 (v/v) is fixed.

No flavour was added to the liquids so as to prevent any interference with the studied phenomenon capable of leading to errors of interpretation.

iv. Chemical products and analytical apparatus used

The target molecules like 3-MCPD and sucralose and the methanol used as solvent during the analyses have analytical quality (purity ≥ 98.0%).

The solutions of PG and VG meet the quality requirements of the European Pharmacopeia (Ph. Eur.) and have purity greater than or equal to 99.5%.

The molecule 3-MCPD was quantified by gas phase chromatography coupled with mass spectrometer: GC-MS (Thermo Trace GC 1300 ISQ). This analysis method was developed within our lab. Thus, a validation dossier was written, which allowed us to define the uncertainty of the method at 22.6%.

The sucralose and nicotine were quantified by ultra-high-performance liquid chromatography coupled with a UV detector and a mass spectrometer: UHPLC-UV/MS (Waters Acquity UPLC H-Class PDA QDA).

The different analytical methods used during this study were qualified for their accuracy, robustness and repeatability.

b. Results

i. Vaporisation of sucralose

Sucralose is a molecule having a relatively important molecular weight. The weight of the molecule (398 g/mol), more than 2 times greater than that of nicotine (162 g/mol), could be a potential brake to efficient vaporisation.

The *Figures 3 and 4* below show the rate of recovery of sucralose in the condensers according to the device used for different concentrations of sucralose and different PG/VG ratios in the liquid.

Overall, the quantities of sucralose recovered in the condensers appear to depend on the chemical composition of the vaporised solution (concentration of sucralose and PG/VG ratio) and different according to the device in question.

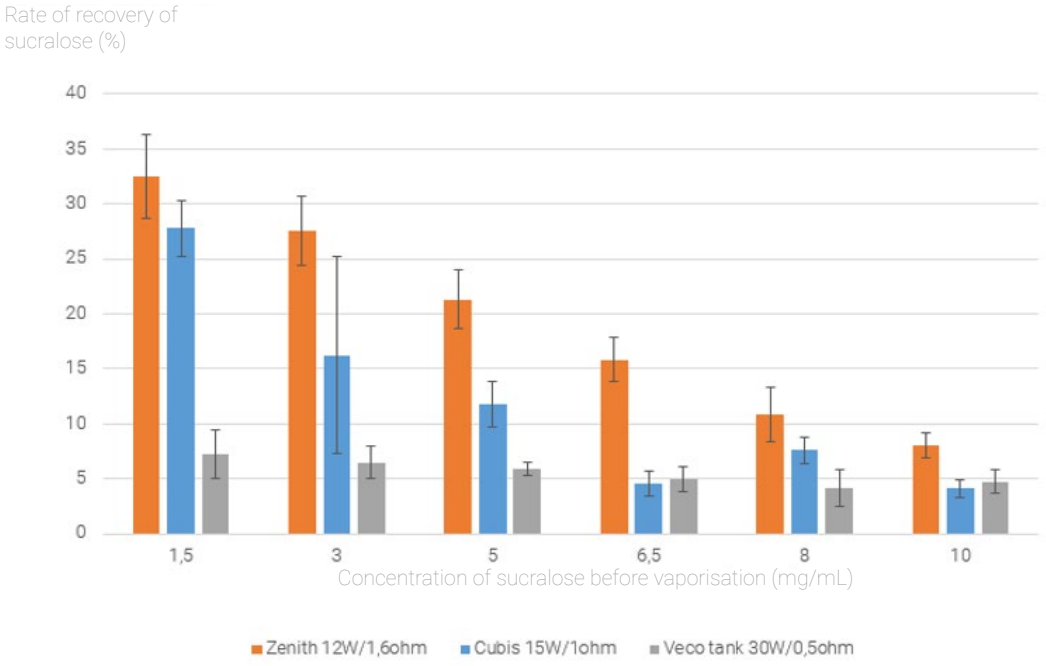

Figure 3: Quantity of sucralose recovered in the aerosol with each vaporisation condition according to the sucralose concentration of the liquid (PG/VG; 20/80 (v/v)) before vaporisation.

Figure 3 shows considerable variability in the recovery of sucralose between the 3 devices tested. These differences are all the greater as the sucralose concentration of the liquid is low.

We observe that for the «Zenith» and «Cubis» devices the lower the initial concentration of sucralose in the solution, the higher its rate of recovery after vaporisation. For the «Veco tank» device the rate of recovery of sucralose varies very little according to the sucralose concentration of the liquid tested.

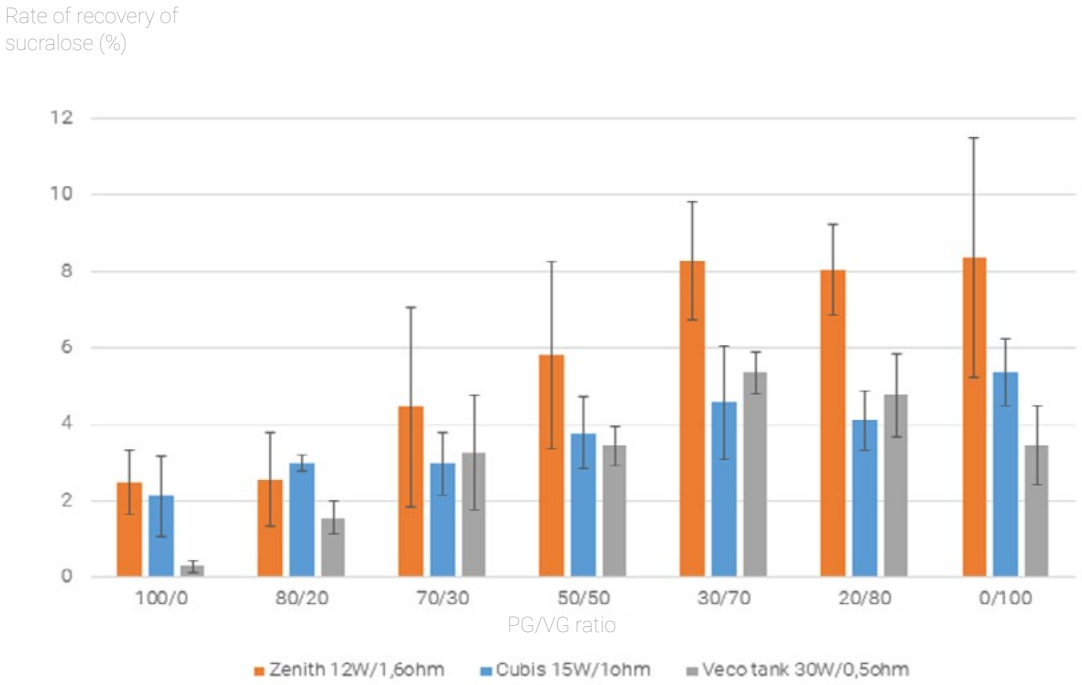

Figure 4: Quantity of sucralose recovered in the aerosol at each vaporisation condition according to the PG/VG ratio of the liquid ([sucralose] = 10 mg/mL) before vaporisation.

Figure 4 tends to show that an increased proportion of VG in the solution favours sucralose recovery in the condenser. However, it is difficult to reach any conclusion on the significance of this phenomenon because of the variability of the measured results.

These measurements highlight the dominance of the device in the efficiency of sucralose vaporisation.

Several hypotheses can then be put forward:

(1)Sucralose does not vaporise correctly and accumulates in the reservoir or in the wick of the heating element.

(2)Sucralose is mostly degraded during vaporisation.

(3) The cryogenic trapping used is not suited to recovery of sucralose.

Hypotheses 1 and 2 remain plausible at this stage of the study and agree with the results available in the scientific literature[22].

Indeed, Rosbrook, K et *al*. (2017) measure only 10% of the quantity of sucralose of the liquid in the latter's vapour. This is with the use of a trapping typology different from that used in the present study.

Thus, hypothesis 3 would appear improbable since similar results are obtained by the use of different trapping systems.

Considered together, these results then suggest poor sucralose vaporisation, high degradation of the molecule or the coexistence of both these phenomena in the conditions tested.

ii. Visual appearance of the wicks and resistance wires

Before studying 3-MCPD production, the lab verified the visual appearance of the heating elements of the devices used after the different operations.

The cotton wicks of the heating elements have a «brown» even «black» appearance and a characteristic burnt odour.

In general, the higher the concentration of sucralose and the proportion of PG in the liquid, the greater this colouring is. In parallel with the colouring of the wicks, the resistances have black particles deposited on the resistance wires.

The colouring of the liquid varies according to the equipment but reproduces the same scheme as obtained with observation of the wicks. The higher the concentrations of sucralose and PG, the stronger is the colouring (Appendix).

These observations confirm the hypothesis according to which sucralose vaporises poorly in the conditions used for performing the operations.

iii. Influence of the PG/VG ratio on 3-MCPD production

The bibliographic summary suggests the formation of 3-MCPD during the vaporisation of sucralose using a vaping device. This phenomenon would be favoured by the presence of VG.

The lab now wishes to verify the production of 3-MCPD during vaporisation of solutions containing 20 mg/mL of nicotine; 10 mg/mL of sucralose and different PG/VG ratios. This is with the 3 devices and resistances selected for carrying out this study.

Figure 5 below shows the quantities of 3-MCPD produced during vaporisation of the liquids for each personal vaporiser:

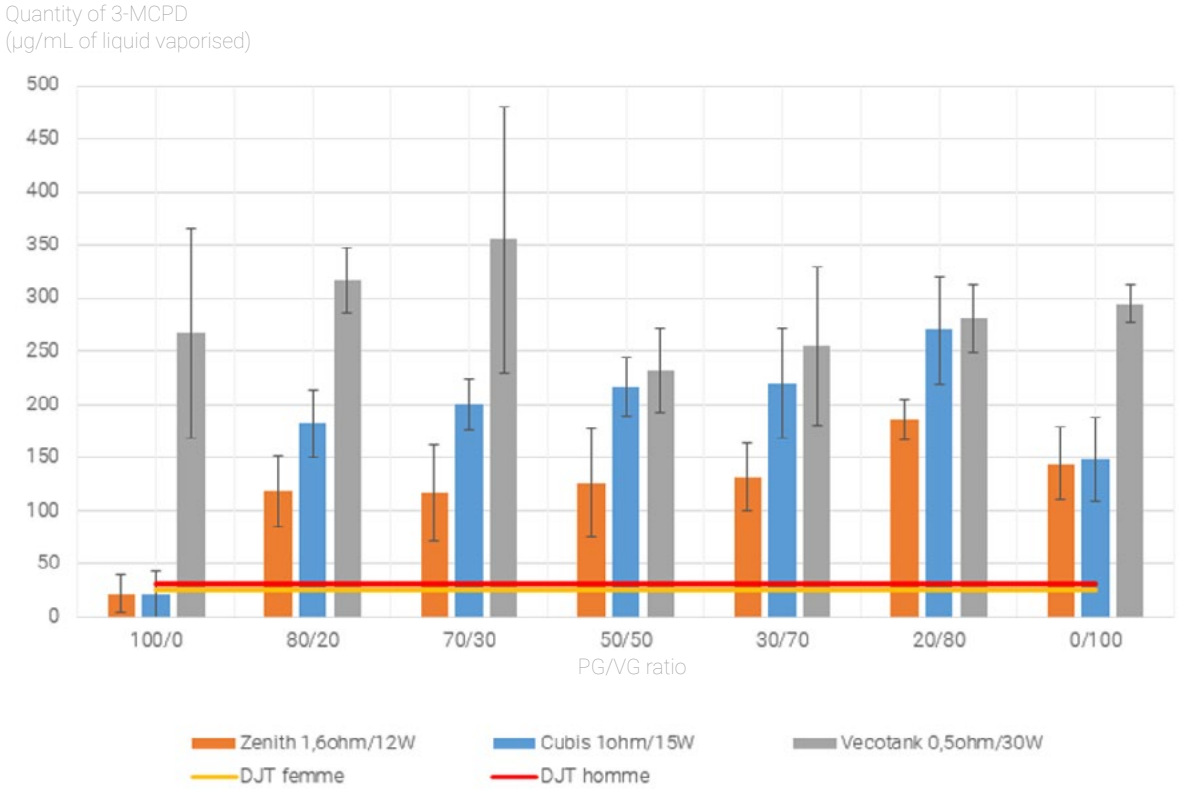

Figure 5: Influence of the PG and VG ratio of a liquid containing sucralose on the production of 3-MCPD during its vaporisation.

Given the results, the PG/VG ratio of the liquid only seems to influence very little the quantity of 3-MCPD generated during its vaporisation apart from those constituted uniquely of PG. This is for all the devices tested.

3-MCPD is detected in the emissions generated in each of the experimental conditions implemented.

Like the rate of recovery, the vaping device considered seems to influence the quantity of 3-MCPD generated during the heating of the liquids. These differences appear more marked between the «Zenith» and the two other devices («Cubis» & «Veco tank»).

iv. Influence of the level of sucralose on 3-MCPD production

In order to have an overall idea of the parameters of influence on the degradation of sucralose into 3-MCPD during its vaporisation using a vaping device, we now wish to study the impact of the sucralose concentration of the liquid on this phenomenon.

Thus, for each of the devices and resistances selected, different solutions containing sucralose concentrations varying from 1.5 to 10 mg/mL were vaporised. All the liquids are made up with 20 mg/mL of nicotine and with a PG/VG ratio of 20/80.

Figure 6 below shows the quantities of 3-MCPD produced during vaporisation of the liquids for each personal vaporiser studied.

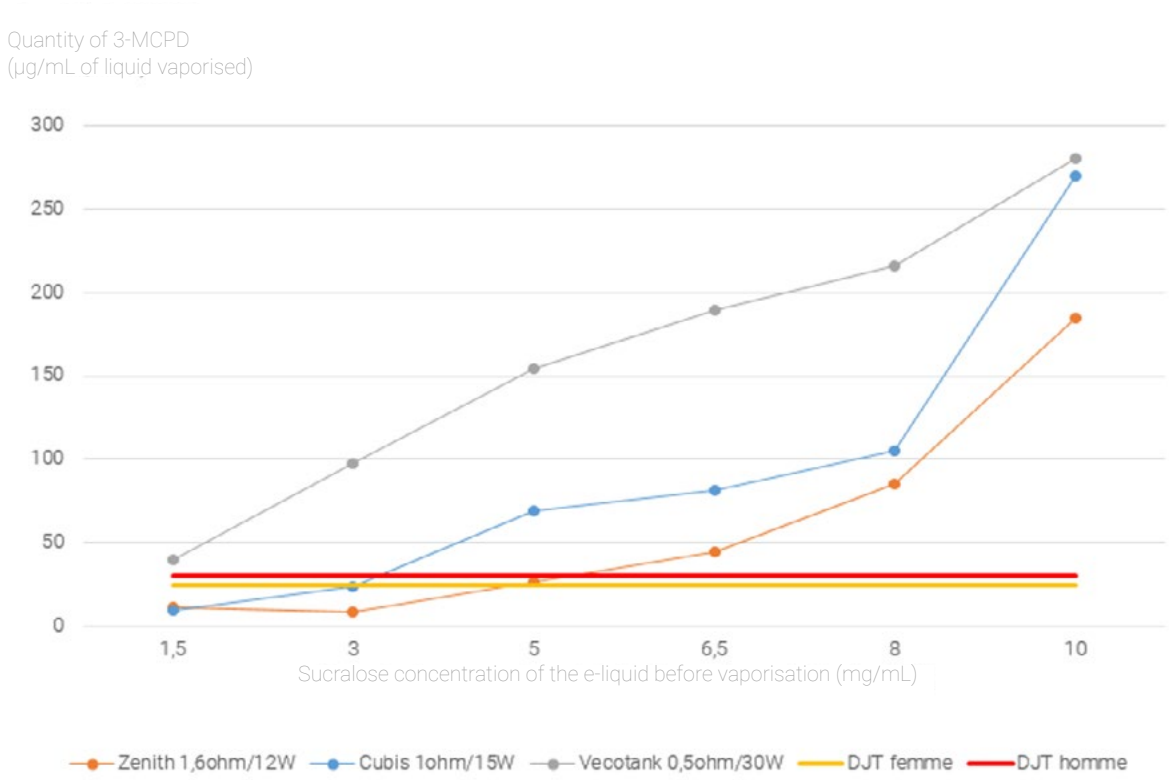

Figure 6: Influence of the level of sucralose in a liquid on the production of 3-MCPD during its vaporisation.

The results show that the presence of sucralose in the liquid induces production of 3-MCPD in its emissions.

In particular it appears that the more the sucralose concentration increases in the liquid to be vaporised, the more the concentration in 3-MCPD increases in the aerosol. This is for all the vaping devices used in this study.

Finally, for each of the sucralose concentrations tested, the production of 3-MCPD during the vaporisation of the liquids varies according to the vaping device considered.

c. Interpretation and discussions

The results presented in this report indicate that the presence of sucralose in a vape liquid induces the formation of 3-MCPD during its vaporisation. This is in all the conditions tested during the study.

The results appear to be influenced by the vaping device considered in conformity with what is suggested by the literature on the subject[22].

The concentration values of 3-MCPD measured during the different experiments should be interpreted in relation to the toxicological aspect of the molecule by the oral (ingestion) and respiratory (inhalation) routes.

i. Toxicological data

To date, too few toxicological studies on 3-MCPD in inhalation have been carried out to make any conclusions on the specific toxicity of the compound by the respiratory route. The great majority of data found concerns exposure by the oral route, i.e. by ingestion.

The CIRC (*Centre International de Recherche sur le Cancer*) concluded that there is sufficient proof of the carcinogenicity of 3-MCPD by ingestion, in animals. According to their overall assessment, 3-MCPD would be likely to be carcinogenic for humans (CMR group 2B), in ingestion[9].

In addition, the expert group of the EFSA (European Food Safety Authority) in 2017 assessed the potential risks of the target molecule in ingestion[26].

The tolerable daily intake (TDI) of 3-MCPD is 2.0 µg/kg (body weight), in ingestion[28].

The TDI for a woman of 63 kg29 is thus 126 µg/d and 154 µg/d for a man of 77 kg by ingestion.

In order to understand the impact that 3-MCPD could have, in inhalation, the lab approached Dr Eric Blouin, toxicological expert with PHYSIOTOX. According to his thorough research[30-34], the toxicity data after chronic exposure by inhalation to 3-MCPD is very little and the acute toxicity data available shows comparable toxic doses by ingestion and inhalation.

Thus, without direct proof of greater toxicity by the respiratory route, the tolerable intake in inhalation will be related to that in ingestion and inhalation, that is: 2 µg/kg per day.

ii. In vaping products

In order to assess the toxicological impact of the use of sucralose in vaping products of and the inhalation of 3-MCPD that it imposes on vapers, the TDI (in µg of 3-MCPD per mL of liquid consumed) were calculated (*Figures 5 & 6*) based on the following argument:

- According to the tolerable daily intake of 3-MCPD in ingestion, a female vaper of 63 kg «could» consume up to 126 µg of 3-MCPD per day while a male vaper of 77 kg «could» consume up to 154 µg of it daily.

- Given an average daily consumption of 5 mL of liquid, a female vaper of 63 kg should then vape so as to inhale a maximum of 25.2 µg of 3-MCPD per mL of liquid consumed in order to follow the TDI of the molecule in ingestion.

- In the same way, a male vaper of 77 kg, consuming 5 mL per day, should vape so as to inhale a maximum of 30.8 µg of 3-MCPD per mL of liquid consumed in order to follow the TDI of the molecule in ingestion.

As *Figures 5 and 6* of this report show, most of the experimental conditions used in this study lead to 3-MCPD generation greater than the theoretical limits.

Most of the quantities of 3-MCPD obtained by the lab then exceed the tolerable daily intake of 3-MCPD as far as to exceed them by a factor of 10 in certain conditions.

It should be noted that the results presented in this report are specific to the conditions employed: chemistry of the liquid and physics of vaporisation. As the liquids do not contain flavours, possible interactions with other chemical compounds traditionally present in vaping products cannot be excluded.

In this study, the lab concentrated uniquely on the production of 3-MCPD during the vaporisation of sucralose. However, it is important to recall that 3-MCPD is only one of many degradation products of sucralose. In order to carry out an overall study on the toxicological consequences related to the use of this sweetener, it would be necessary to study all of its specific degradation products and its influence on the degradation phenomena traditionally found in the vaping context.

Finally, to date no exposure limit value of 3-MCPD exists in inhalation since so far there is no case of chronic exposure. However, it is highly probable that the toxicological risk of the molecule in inhalation is at least as great as in ingestion.

4. CONCLUSION

In response to the demand of the market for sweetened products, certain manufacturers of vaping liquids have incorporated sucralose in the formulation of their products.

Analysis of the scientific literature was required to understand the thermal degradation of sucralose, identify its main degradation products as well as their toxicological profile.

Following this bibliographic summary, Ingésciences wished to carry out an experimental study in order to estimate the toxicological issues related to its use in vaping products.

This showed that very little sucralose is transmitted to the vaper during vaporisation of a liquid containing it. The study also showed the degradation of sucralose into 3-MCPD and after vaporisation using three separate devices.

The vaping device considered and the sucralose concentration of the vaping product influence these phenomena particularly.

3-MCPD is recognised as CMR (group 2B) at least in ingestion. Its regular inhalation above the tolerable daily intake in ingestion could represent a risk for the consumer.

While the literature indicates several products of thermal degradation related to sucralose, only 3-MCPD was studied by the lab. And the list of the degradation molecules of sucralose contains compounds recognised as toxic in inhalation (such as 1,2 & 1,3-chloropropanols[19,20]).

The consumption of a liquid containing sucralose thus entails a greater toxicological risk than that of a product free from it.

The vaping products industry is already aware of these issues and its attention is currently drawn to other alternative sweeteners. Unlike sucralose, the thermal stability and the toxicological impact of the use of these compounds should be studied before their incorporation in a vaping liquid.

5. BIBLIOGRAPHIC REFERENCE

1- Morean, M. E., Butler, E. R., Bold, K. W., Kong, G., Camenga, D. R., Cavallo, D. A., ... & Krishnan-Sarin, S. (2018). Preferring more e-cigarette flavors is associated with e-cigarette use frequency among adolescents but not adults. *PloS one, 13*(1), e0189015.

2- Schiffman, S. S., & Rother, K. I. (2013). Sucralose, a synthetic organochlorine sweetener: overview of biological issues. *Journal of Toxicology and Environmental Health, Part B, 16*(7), 399-451.

3- Schiffman, S. S., Sattely-Miller, E. A., & Bishay, I. E. (2008). Sensory properties of neotame: Comparison with other sweeteners.

4- Wiet, S. G., & BEYTS, P. K. (1992). Sensory characteristics of sucralose and other high intensity sweeteners. *Journal of Food Science, 57*(4), 1014-1019.

5- AFNOR XP D90-300-2- Cigarettes électronique et e-liquides – Partie 2 : exigences et méthodes d'essais relatives aux cigarettes e-liquides.

6- Grotz, V. Lee, and Ian C. Munro. «An overview of the safety of sucralose.» *Regulatory toxicology and pharmacology* 55.1 (2009): 1-5.

7- U.S. Food and Drug Administration. 1998. Food additives permitted for direct addition to food for human consumption; sucralose. 21CFR Part 172 [Docket No. 87F-0086]. *Fed. Reg. 63*(64): 16417–16433.

8- European Union. 2004. Directive 2003/115/EC of the European Parliament and of the Council of 22 December 2003 amending Directive 94/35/EC on sweeteners for use in foodstuffs. *Off. J. Eur. Union* 47 (L24), 65–71.

9- Mace, Oliver J., et al. «Sweet taste receptors in rat small intestine stimulate glucose absorption through apical GLUT2.» *The Journal of physiology* 582.1 (2007): 379-392.

10- Mace, Oliver J., et al. «An energy supply network of nutrient absorption coordinated by calcium and T1R taste receptors in rat small intestine.» *The Journal of physiology* 587.1 (2009): 195-210.

11- Abou-Donia, Mohamed B., et al. «Splenda alters gut microflora and increases intestinal p-glycoprotein and cytochrome p-450 in male rats.» *Journal of Toxicology and Environmental Health, Part A* 71.21 (2008): 1415-1429.

12- Roberts, A., et al. «Sucralose metabolism and pharmacokinetics in man.» *Food and chemical toxicology* 38 (2000): 31-41.

13- Brown, Rebecca J., and Kristina I. Rother. «Non-nutritive sweeteners and their role in the gastrointestinal tract.» *The Journal of Clinical Endocrinology & Metabolism* 97.8 (2012): 2597-2605.

14- Barndt, R. L., and G. Jackson. «Stability of sucralose in baked goods.» *Food Technology (Chicago)* 44.1 (1990): 62-66.

15- Parkin, Kirk Lindsay, Owen R. Fennema, and Srinivasan Damodaran, eds. *Fennema's food chemistry*. CRC Press, 2008.

16- World Health Organization. 2002. 3-Chloro-1,2-propanediol. In *Safety evaluation of certain food additives and contaminants*. WHO Food Additives Series 48.

17- Hutchinson, Sheryl A., Gregory S. Ho, and Chi-Tang Ho. «Stability and degradation of the high-intensity sweeteners: Aspartame, Alitame, and Sucralose.» *Food reviews international*15.2 (1999): 249-261.

18- Bannach, Gilbert, et al. «Thermal stability and thermal decomposition of sucralose.» *Eclética Química* 34.4 (2009): 21-26.

19- A Rahn, VA Yaylayan - Thermal degradation of sucralose and its potential in generating chloropropanols in the presence of glycerol – Food Chemistry, 2010.

20- De Oliveira, Diogo N., Maico De Menezes, and Rodrigo R. Catharino. «Thermal degradation of sucralose: a combination of analytical methods to determine stability and chlorinated byproducts.» *Scientific reports* 5 (2015): 9598.

21- Duell, Anna K., et al. «Boiling points of the propylene glycol+ glycerol system at 1 atmosphere pressure: 188.6–292° C without and with added water or nicotine.» Chemical engineering communications 205.12 (2018): 1691-1700.

22- Rosbrook, K., Erythropel, H. C., DeWinter, T. M., Falinski, M., O'Malley, S., Krishnan-Sarin, S., ... & Green, B. G. (2017). The effect of sucralose on flavor sweetness in electronic cigarettes varies between delivery devices. *PloS one*, 12(10).

23- Duell, A. K., McWhirter, K. J., Korzun, T., Strongin, R. M., & Peyton, D. H. (2019). Sucralose-enhanced degradation of electronic cigarette liquids during Vaping. *Chemical research in toxicology*, 32(6), 1241-1249.

24- El-Hage, R., El-Hellani, A., Haddad, C., Salman, R., Talih, S., Shihadeh, A., ... & Aoun Saliba, N. (2019). Toxic emissions resulting from sucralose added to electronic cigarette liquids. *Aerosol Science and Technology*, 53(10), 1197-1203.

25- Andres, Susanne, Klaus E. Appel, and Alfonso Lampen. «Toxicology, occurrence and risk characterisation of the chloropropanols in food: 2-monochloro-1, 3-propanediol, 1, 3-dichloro-2-propanol and 2, 3-dichloro-1-propanol.» *Food and Chemical Toxicology* 58 (2013): 467-478.

26- IARC: International Agency for Research on Cancer. 3-Monochloro-1,2-propanediol (updated 14 September 2016). In: IARC Monographs. Volume 101. 2013. Some Chemicals Present in Industrial and Consumer Products, Food and Drinking-water. Lyon, France, 349-74.
https://monographs.iarc.fr/wp-content/uploads/2018/06/mono101-010.pdf

27- AFNOR XP D90-300-3- Cigarettes électroniques et e-liquides – Partie 3: exigences et méthodes d'essai relatives aux émissions

28- Article : « Mise à jour du niveau de sécurité pour le 3-MCPD dans les huiles végétales et les aliments », European Food Safety Authority (EFSA), 2018

29- Thibaut de Saint Pol, division Conditions de vie des ménages, Insee (Institut national de la statistique et des études économiques), L'obésité en France : les écarts entre catégories sociales s'accroissent, INSEE PREMIERE, 2007, N°1123

30- Sunahara G, Perrin I, Marchesini M. 1993. Carcinogenicity study on 3-monochloro-1,2-propanediol (3-MCPD) administered in drinking water to Fischer 344 rats. Unpublished report No. RESR93003. Nestec Ltd. Research & Development, Switzerland. Cited by EFSA, 2018.

31- Robjohns S, Marshall R, Fellows M, Kowalczyk G. In vivo genotoxicity studies with 3-monochloropropan-1,2-diol. Mutagenesis. 2003. 18:401-4.

32- Onami S, Cho YM, Toyoda T, Horibata K, Ishii Y, Umemura T, Honma M, Nohmi T, Nishikawa A, Ogawa K. Absence of in vivo genotoxicity of 3-monochloropropane-1,2-diol and associated fatty acid esters in a 4-week comprehensive toxicity study using F344 gpt delta rats. Mutagenesis. 2014. 29:295-302.

33- Lee BS, Park SJ, Kim YB, Han JS, Jeong EJ, Son HY, Moon KS. Twenty-six-week oral carcinogenicity study of 3-monochloropropane-1,2-diol in CB6F1-rasH2 transgenic mice. Arch Toxicol. 2017. 91(1):453-64.

34- Lee BQ, SM Khor. 3-Chloropropane-1,2-diol (3-MCPD) in Soy Sauce: A Review on the Formation, Reduction, and Detection of This Potential Carcinogen. 2015. 14(1): 48-66.

6. APPENDIX

a. Vaporisation using the clearomizer, CUBIS 15 W, 1 Ω

Figure 7: Wicks of resistances obtained during the vaporisation of e-liquids (PG/VG 20/80), nicotine at 20 mg/mL and with different levels of sucralose with the CUBIS clearomizer. From left to right, the sucralose concentrations are 1.6, 2.3, 4.6, 8.7, 10 mg/mL.

Figure 8: Wicks of resistances obtained during the vaporisation of e-liquids with different PG/VG ratios and each containing 20 mg/mL nicotine and 10 mg/mL sucralose with the CUBIS clearomizer. From left to right, the PG/VG ratios are: 100% PG; 80/20; 70/30; 50/50; 30/70; 20/80; 100% VG.

b. Vaporisation using the clearomizer, Zenith 12 W, 1.6 Ω

Figure 9: Wicks of resistances obtained during the vaporisation of e-liquids (PG/VG 20/80), nicotine at 20 mg/mL and with different levels of sucralose with the Zenith clearomizer. From left to right, the sucralose concentrations are 1.5, 3, 5, 6.5, 8, 10 mg/mL.

Figure 10: Wicks of resistances obtained during the vaporisation of e-liquids with different PG/VG ratios and each containing 20 mg/mL nicotine and 10 mg/mL sucralose with the Zenith clearomizer. From left to right, the PG/VG ratios are: 100% PG; 80/20; 70/30; 50/50; 30/70; 20/80; 100% VG.

c. Vaporisation using the clearomizer, Veco tank 30 W, 0.5 Ω

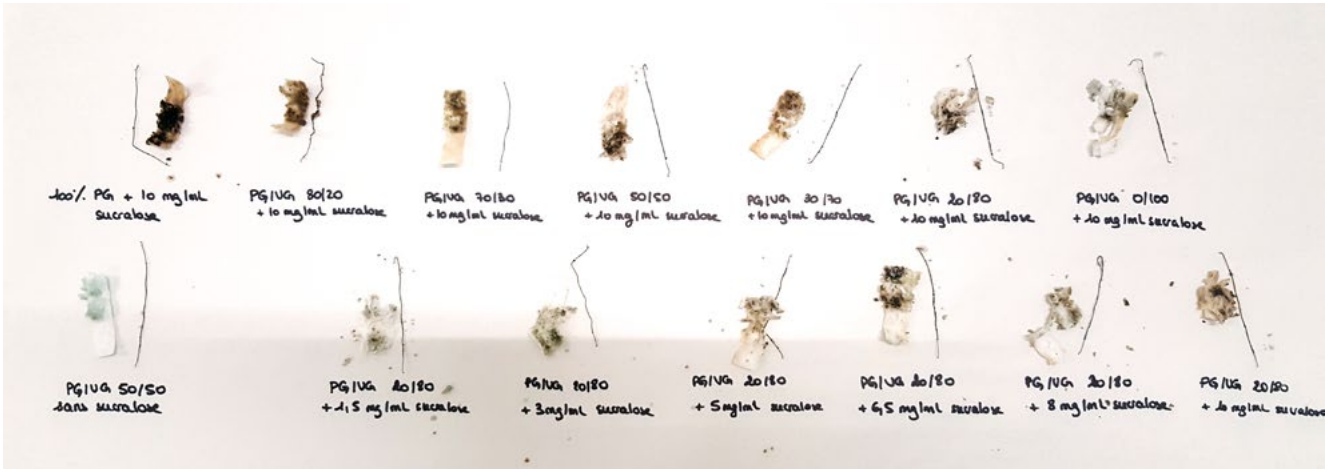

Figure 11: (Top) Wicks of resistances obtained during the vaporisation of e-liquids with different PG/VG ratios and each containing 20 mg/mL nicotine and 10 mg/mL sucralose with the Zenith clearomizer. From left to right, the PG/VG ratios are: 100% PG; 80/20; 70/30; 50/50; 30/70; 20/80; 100% VG.

(Bottom) Wicks of resistances obtained during the vaporisation of e-liquids (PG/VG 20/80), nicotine at 20 mg/mL and with different levels of sucralose with the Veco tank clearomizer. From left to right, the sucralose concentrations are 0, 1.5, 3, 5, 6.5, 8, 10 mg/mL. (The e-liquid not containing sucralose has PG/VG ratio at 50/50).