DRUGS WORKING GROUP

CALCULATOR FOR			
SAMPLING FOR QUANTITATIVE ANALYSIS			
VALIDATION REPORT			
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1. INTRODUCTION

A representative sampling procedure can be performed on a population of units with sufficient similar external characteristics (e.g. granularity, colour). Different sampling approaches, i.e. arbitrary or statistical, may be applied and the *incremental sampling* approach is one of them. This approach allows the calculation of an appropriate number of increments and the weights of primary sample (m_1) and analytical sample (m_n), depending on the accepted sampling uncertainty, which can be estimated by the fundamental error (s_{FE}) of the sampling process.

This document refers to the Excel based "ENFSI - Drugs Working Group (DWG) Calculator for Sampling of Illicit Drugs for Quantitative Analysis".
The theoretical background of the applied sampling protocol and the correctness of the deduced formula for

- appropriate number of increments (n)
- weight of analytical sample (m_n)

has been published [1],[2],[3] and summarized in the ENFSI DWG Guidelines on Sampling of Illicit Drugs for Quantitative Analysis[4].
This documents validates the correct input and output by Excel of all needed parameters.

2. DEFINITIONS

Some definitions and labels as applied in this document:

s_{FE} Fundamental error, expressed as the RSD of the sampling process. The fundamental error (FE) is the minimum error generated when a sample of a given weight is collected and is influenced by its particle size. The fundamental error is inherent in the compositional distribution of the sample material and cannot be changed without changing the nature of the sample. This error can be reduced, either by increasing the mass

of the sample or by decreasing the size of the largest particles in it, by comminution.

a_L The real (but unknown and to be evaluated) mass fraction (w/w) of the analyte (of a certain lot).

In the calculator it is used as minimum assumed ratio (w/w) of target analyte to matrix.

d Nominal particle size in the sample in cm – experimental values are given in the calculator in μm unit. Nominal particle size is equivalent to the maximum particle size in the lot to be sampled. In practice, d is taken to be the screen size that retains 5% of the lot being sampled.

m_1 Primary sample in g - chosen material obtained from the bulk by correct incremental sampling

m_n Analytical sample in g - last chosen material after homogenisation and mass reduction, which will be dissolved for instrumental analysis. The calculator converts the value to mg unit.

n Number of increments for assembling the primary sample - in the calculator the increment size is fixed to 1 g of powder or 1 tablet or 1 cannabis bud.

s_{1g} Typical heterogeneity at 1 g level.

s_{nx1g} Maximum acceptable heterogeneity (as % RSD) after the sampling process – usually close to the RSD of the analytical method.

3. FORMULA USED IN THE CALCULATOR

$$m_n = \frac{0.125\left(\frac{1-\left(0.7a_L+0.3a_L^2\right)}{a_L}\right)d^3}{s_{FE}^2}$$

Equation 5 in the Guidelines Literature [4]. The equation is solved to m_n, the weight of the analytical sample

$$n = \left(\frac{s_{1g}}{s_{nx1g}}\right)^2$$

Equation 9 in the Guidelines Literature [4]. The equation is solved to n, the number of increments.

4. SOFTWARE

The validation has been performed with Excel® 2003 and 2013.

5. VALIDATION

5.1. Data required for calculation (input)

The calculator only allows typing, where an input value is needed. All other fields, especially those, which contain formulas and calculation are protected.

The typical heterogeneity of powders, Cannabis resin, Cannabis buds and 'Ecstasy' tablets are given using the values published in literature [1,2,3]. However, the user can choose another heterogeneity, if appropriate values or examinations are available. This value is to be set in Cell C9. In the same way, the size of the increment and the nominal particle size can be chosen in Cell E9 and F9. These cells are enabled and change to yellow fields, when the **type of material** in Cell C16 is set to option *5. Other*. Therefore **Cell C16 is the starting point for using the calculator**. The selection in C16 will influence the values set in C18, E18 and C21, as they are taken from the table above. The selection is correct.

If *5. Other* as type of material is used, the input value for the **typical heterogeneity** is limited within the range of 1 to 70. Heterogeneity of 70% is the uppermost limit, as it reflects the unrealistic situation of having one part of the material with 100% concentration (pure target compound) and the remaining part of the material with 0% concentration (only diluting or 'cutting' agents). This extreme situation, within one material, would result in a heterogeneity of approximately 70%. However, this situation is quite unrealistic. The same maximum overall heterogeneity would arise, if a seizure consisted of 2 separate visually undistinguishable packages, where one contains the pure compound and the other only diluting or cutting agents.
The guidelines and sampling plan are based on situations where multiple package samples are divided into sub-samples, until the sub-samples are visually undistinguishable or other reasons exist to consider them as belonging to the same group.

The default value for the **increment level** in Cell E9 is set to 1 g. This value is flexible and can be chosen by the user. However, when other increment levels are chosen, the user has to know the corresponding fundamental error s_{FE} , expressed as the RSD_{sampl} from their own experiments or from the literature. The text field will be repeated in Cell E18, G30 and D44. The links are checked and correct.

Allowed values in cell F9, the **nominal particle size of the comminuted material,** are within the range of 10 μm to 5000 μm

The **estimated heterogeneity** in cell C18 will be filled automatically, depending on the chosen type of material. The links are checked and correct.

The **increment level** in cell E18 will be filled automatically, depending on the chosen type of material. The links are checked and correct.

The **estimated particle size** of the comminuted material in cell C21 will be filled automatically, depending on the chosen type of material. The links are checked and correct.

The **Maximum intended uncertainty** in cell C24 (arising from the sampling process and related to the fundamental error) has to be chosen in connection with the laboratory's uncertainty of the analytical process (RSD_{sampl}).
It should be noted that, the variance of the fundamental error s^2_{FE} is needed in the formula given in chapter 3 (Equation 5). This means that the chosen RSD_{sampl} in cell C24 - considered as s_{FE} – has to be squared in the formula.
The range is limited between 0.1 % and the value set in cell C18, which is the selected value of estimated heterogeneity. A higher value is not reasonable.
The value of cell C24 is used in 2 equations, calculated in the minimum number of increments (n), cell E31 and the minimum analytical sample weight m_n, cell E34.

The **Minimum estimated concentration of the materials** in cell C27 is limited to the range of 0.1% and 100%. This value is used in the calculation of the minimum analytical sample weight m_n, cell E34.

5.2. **Calculated data (output)**

The calculator has two output values

  - Minimum number of increments in cell E31 and
  - Minimum analytical sample weight in cell E34.

Minimum number of increments is based on Eq. 9 from the guidelines.

Minimum analytical sample weight is based on Eq. 5 from the guidelines.

The formulas are correct as published in the guidelines [4] and literature [1,2,3].

The calculated numbers are always rounded up to the next integer value.

The calculated result in cell E31 is correctly repeated in cell C44.

The calculated result in cell E34 is correctly repeated in D39 and D49.

In the following chapters 5.3. and 5.4. a series of normal (to be expected in casework) and extreme (non-realistic) values are calculated by hand with a pocket calculator and compared to the results calculated by the Excel software.

5.3. **Calculation of the number of increments**

At the stage of taking the primary sample, only the heterogeneity of the untreated material and the desired sampling uncertainty is relevant. The particle size is not relevant and is taken into consideration, when the weight of the analytical sample (see chapter 5.4) is calculated.

Heterogeneity of the material s_{1g} [%] (Input value in cell C9)	Maximum intended uncertainty $s_{n\times1g}$ [%] (Input value in cell C24)	Number of increments calculated by pocket calculator $n = \left(\dfrac{s_{1g}}{s_{nx1g}}\right)^2$	Number of increments calculated by Excel software $n = \left(\dfrac{s_{1g}}{s_{nx1g}}\right)^2$ (Output value in cell C31)
1	0.1	100	100
	0.5	4	4
	1	1	1
5	0.1	2,500	2,500
	1	25	25
	2	6.25	7
	5	1	1
10	0.1	10,000	10,000
	1	100	100
	5	4	4
	10	1	1
20	0.1	40,000	40,000
	1	400	400
	5	16	16
	10	4	4
	15	1.78	2
40	0.1	160,000	160,000
	1	1,600	1,600
	5	64	64
	10	16	16
	20	4	4
70	0.1	490,000	490,000
	1	4,900	4,900
	10	49	49
	20	12.25	13
	30	5.44	6

In practice, a Maximum intended uncertainty $s_{n \times 1g}$ for chromatographic methods of lower than 1% is, even for extremely homogenised material used for proficiency tests, not realistic and also would lead to an unreasonable number of increments being taken. In general, high heterogeneities of the untreated material in its original state should lead to suitable action. For example a multi-unit seizure might be split into sub-groups. A huge one-unit seizure, with a very high heterogeneity, needs special consideration, like increasing the number of increments to ensure a narrow result range, in duplicate sampling or other sampling approaches. See table 24B in the guidelines.

The calculation by hand using a pocket calculator and the use of the Excel calculator led to the same results. The Excel calculator is rounding up to the next integer value.

5.4. **Calculation of the minimum analytical sample weight**

ratio (w/w) of target analyte to matrix a_L (Input value in cell C27)	nominal particle size d in cm (Input value in cell F9)	Maximum intended uncertainty s_{FE} (Input value in cell C24)	weight of the analytical sample in mg calculated by pocket calculator $m_n = \dfrac{0.125 \left(\dfrac{1 - (0.7a_L + 0.3a_L^2)}{a_L} \right) d^3}{s_{FE}^2}$	weight of the analytical sample in mg calculated by Excel software (Output value in cell E34)
0.001 (0.1%)	0.001 (10 um)	0.001 (0.1%) #	124.913	125
		0.05 (5%)	0.05	1
		0.2 (20%)	0.004	1
	0.01 (100 um)	0.001 (0.1%) #	124912.5 #	124913
		0.05 (5%)	49.965	50
		0.2 (20%)	3.123	4
	0.1 cm (1000 um)	0.001 (0.1%) #	1.25E+08 #	124912463
		0.05 (5%)	49964.99#	49965
		0.2 (20%)	3122.812	3123

0.1 (10%)	0.001 (10 um)	0.001 (0.1%) #	1.159	2
		0.05 (5%)	0.001	1
		0.2 (20%)	0.001	1
	0.01 (100 um)	0.001 (0.1%) #	1158.75	1159
		0.05 (5%)	0.464	1
		0.2 (20%)	0.029	1
	0.1 cm (1000 um)	0.001 (0.1%) #	1158750 #	1158750
		0.05 (5%)	463.5	464
		0.2 (20%)	28.969	29
0.5 (50%)	0.001 (10 um)	0.001 (0.1%) #	0.144	1
		0.05 (5%)	0.001	1
		0.2 (20%)	0.001	1
	0.01 (100 um)	0.001 (0.1%) #	143.75	144
		0.05 (5%)	0.058	1
		0.2 (20%)	0.004	1
	0.1 cm (1000 um)	0.001 (0.1%) #	143750#	143750
		0.05 (5%)	57.5	58
		0.2 (20%)	3.594	4

Analytical samples consisting of an unrealistic amount of material are marked with #. In practice the laboratory is in favour of reducing the weight of analytical sample, by either reducing the particle size or accepting higher values for the maximum intended uncertainty.

The calculation by hand using a pocket calculator and the use of the Excel calculator led to the same correct results. The Excel calculator is rounding up to the next mg integer value.

5.4.1. Protection of the software

The Calculator is password protected. Users may only enter data in specific required cells. The password is known to the authors.

6. CONCLUSION

6.1. **Software**

The ENFSI DWG Calculator for Sampling of Illicit Drugs for Quantitative Analysis version 1.1 is validated and fit for purpose.

Input data has a reasonable range of possible values. However, a combination of extreme values could lead to unrealistic output data, i.e. an excessive number of increments or an excessive weight of the analytical sample. It is up to individual laboratories to set data value limits that will provide a reasonable sampling plan.

7. CONTACT ADDRESS

Please address questions, report errors and/or bugs found in the Excel software or within this document to e-mail: csesztregi.tamas@orfk.police.hu , michael.bovens@for-zh.ch or to the DWG contact person through the contact form (http://www.enfsi.eu/about-enfsi/structure/working-groups/drugs).

Tamas Csesztregi

e-mail: csesztregi.tamas@orfk.police.hu

Head of Chemistry Department

Institute for Forensic Sciences,

Mosonyi 9

1089 Budapest

Hungary

Dr. Michael Bovens

e-mail : michael.bovens@for-zh.ch

Chief Scientist

Forensic Science Service Zurich

PO Box

8021 Zurich

Switzerland

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