

Sample Pooling Calculator (SPoC) – worked example:

1. Enter the project name, unique Sample Donor ID and the number of samples in the appropriate fields.
The final volume of the pooled sample is set to 1ml but this can be changed to any volume (in mls) as needed.
2. Press the “GO” button to generate a list.

Project Name	Example Project
Sample Donor ID	XX01
Number of Samples	10
Final Volume (ml)	1

GO 

Reset Form

Extend sample list

Export and Save

SPoC 

UNIVERSITY OF Southampton

Instructions

- 1) Type your project name and input the sample donor ID and the number of samples you have in the input box to the left.
- 2) The final volume is set to 1ml. To change this, type a different volume (in ml) in the box.
- 3) Press "GO" to generate a table.
- 4) Enter each sample date in the white 'Date' column provided. The values in the other columns will auto-populate.
- 5) If you need to change the final volume, type in a new value in cell M5 and press Enter on your keyboard.
- 6) If you want to start over, press "Reset Form".
- 7) Once you are finished, press "Export and Save". You will be prompted to save the file with a specified name and location.
- 8) Use the calculated volumes (in μ l) to add the correct volume of each sample to your pooled sample.

Created by: Charlotte Stuart, Mark Weal & Ian Galea



3. Next enter the date of each sample in the fields provided.

Note that you can increase the number of samples at any time by clicking the button "Extend sample list".

No.	Sample Donor ID	Date	Sample Day
1	XX01	01/01/2019	0
2	XX01	03/01/2019	2
3	XX01	04/01/2019	0
4	XX01	Enter Date	0
5	XX01	Please enter the date as dd/mm/yy	0
6	XX01		0
7	XX01		0
8	XX01		0
9	XX01		0
10	XX01		0

Unique sample identifier	µL to add
XX01_d0	500
XX01_d2	0
XX01_d0	0

Project Name	Example Project
Sample Donor ID	XX01
Number of Samples	10
Final Volume (ml)	1

GO

[Reset Form](#)

[Extend sample list](#)

[Export and Save](#)

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- 8) Use the calculated volumes (in µl) to add the correct volume of each sample to your pooled sample.

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4. Once all the sample dates have been inputted, click the "Export and Save" button.

No.	Sample Donor ID	Date	Sample Day
1	XX01	01/01/2019	0
2	XX01	03/01/2019	2
3	XX01	04/01/2019	3
4	XX01	05/01/2019	4
5	XX01	06/01/2019	5
6	XX01	08/01/2019	7
7	XX01	10/01/2019	9
8	XX01	11/01/2019	10
9	XX01	12/01/2019	11
10	XX01	15/01/2019	14

Unique sample identifier	µL to add
XX01_d0	71
XX01_d2	107
XX01_d3	71
XX01_d4	71
XX01_d5	107
XX01_d7	143
XX01_d9	107
XX01_d10	71
XX01_d11	143
XX01_d14	107

Project Name	Example Project
Sample Donor ID	XX01
Number of Samples	10
Final Volume (ml)	1

GO

[Reset Form](#)

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[Export and Save](#) 

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5. The sample list opens in a new Excel workbook and the "Save As" dialogue box pops up. Save as a new excel file in the desired location.

Book1 - Excel

File Home Insert Page Layout Formulas Data Review View Developer Add-ins ChemOffice16 ACROBAT Team Tell me what you want to do...

Clipboard Font Alignment Number Styles Cells Ed

A B C D E F G H I J K L M N O P Q

1 Date Sample Day Unique sample identifier μL to add

2

3 01/01/2019 0 XX01_d0 71

4 03/01/2019 2 XX01_d2 107

5 04/01/2019 3 XX01_d3 71

6 05/01/2019 4 XX01_d4 71

7 06/01/2019 5 XX01_d5 107

8 08/01/2019 7 XX01_d7 143

9 10/01/2019 9 XX01_d9 107

10 11/01/2019 10 XX01_d10 71

11 12/01/2019 11 XX01_d11 143

12 15/01/2019 14 XX01_d14 107

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Save As

File name: Example 1

Save as type: Excel Workbook (*.xlsx)

Authors: Stuart C. Title: Add a title

Tags: Add a tag Subject: Specify the subject

Manager: Specify the manager

Company: University Of Southampt...

Save Cancel

6. Column E shows the calculated volume of each sample. These volumes need to be pooled together to make up a single pooled sample.

This pooled sample should be analysed in triplicate and represents an average of all the samples over the time period selected.

Methodology

Definitions:

1. In a study of n days (d), the study runs from d_0 to d_n . In our example above, the study runs from d_0 to d_{14}
2. s_i = sample
3. s_i is the sample number, where $i = 1 \rightarrow z$, so that $s_1, s_2, s_3 \dots s_z$, where i = the position of a value in a row vector containing the days the samples were taken (d_i). If samples are taken less frequently than daily, $z < n$, and i will be discontinuous. For instance in our example above, the row vector contains the following d_i values: 0, 2, 3, 4, 5, 7, 9, 10, 11, 14. In our example above, z is 10, $n=14$.
4. V is the final volume of the pooled sample. In our example above, $V=1\text{ml}$
5. v_i is the volume that sample s_i will contribute to V , so that $V = v_1 + v_2 + v_3 \dots v_z$. In our example above, $V = 71 + 107 + 71 + 107 + 143 + 107 + 71 + 143 + 107\mu\text{l} = 1\text{ml}$

Algorithm:

The aim is to replace analysis of multiple individual samples with analysis of a single pooled sample. Hence the relative volume contribution of each individual sample to the single pooled sample needs to be proportional to the time interval between the individual sample and its neighbours.

1. Each sample was assumed to reflect the UNCR of:
 - a) the day it was taken, ie d_i
 - b) half the inter-sample time interval preceding day i , i.e. $d_i - d_{(i-1)}$
 - c) half the inter-sample time interval following day i , i.e. $d_{(i+1)} - d_i$

Hence the UNCR of sample s_i is assumed to reflect the UNCR of the time period surrounding it, i.e.

$$t_i = \frac{d_{(i+1 \rightarrow z)} - d_{(i-1)}}{2}$$

See figure across. For example, for sample s_0 , $v_0 = (2-0)/2 = 1$ day; for sample s_5 , $v_5 = (7-4)/2 = 1.5$ days, for sample s_{10} , $v_{10} = (14-11)/2 = 1.5$ days.

2. The contribution of sample s_i to V , i.e. v_i , is this time period t_i divided by the total time period (t_n in days), multiplied by V in μl , so

$$v_i = \frac{t_i}{n} / (1000 \times V)$$

