The Importance of Concentration Factor in SARS-CoV-2 Wastewater Epidemiology Methods

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Concentration Factor is a single factor that provides a direct comparison of sample preparation/concentration methods and their impact on method detection limit – taking into account:

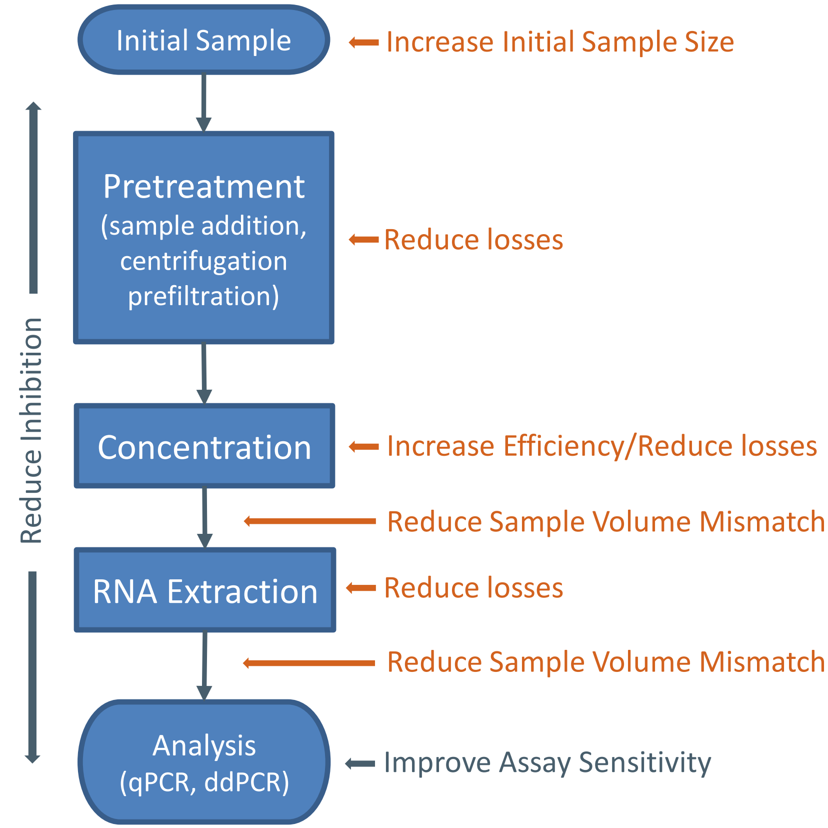
- Initial Sample Volume
- Efficiency of each step
- Volume mismatch between steps

CF = Concentration Factor
$V_{Initial}$ = Volume of Initial Sample (mL)
V_{Final} = Volume of Final Sample (mL)
E_{Method} = Combined Efficiency of Entire Method (%)*
**includes losses due to volume mismatch between steps*

$$CF = \frac{V_{Initial}}{V_{Final}} x E_{Method}$$

CF = Concentration Factor
T_{Final} = Titer of the Final Sample (copies/mL)
$T_{Initial}$ = Titer of the Initial Sample (copies/mL)

$$CF = \frac{T_{Final}}{T_{Initial}}$$



Takeaway: A 50% reduction in initial volume, 50% efficiency or a loss of 50% of sample due to volume mismatch between steps all impact CF and ultimately the method detection limit equally.

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INNOVAPREP

CALCULATING YOUR POTENTIAL **CONCENTRATION FACTOR**

Concentrating Pipette Select™

INTRODUCTION

The primary purpose of the Concentrating Pipette Select system is for the concentration of particles. The CP Select has the ability to remove all of the liquid from the sample, capturing the particles of interest, then delivering the captured particles into volumes of clean buffer as small as 150 microliters.

COMMON TARGET PARTICLES

- Bacteria
- Protozoa
- Fungi
- Molds
- Parasites
- Pollens
- Virus
- Proteins

Common methods for sample concentration include culture-based enrichment, serial filtration steps by manual methods, and centrifugation. All are time, labor, and material intensive.

The Concentrating Pipette has the ability to deliver those particles into an extremely small final volume to better match the input volume of detection methods such as immunoassay, qPCR etc., thus enabling detection at previously undetectable levels. The automated one-pass system rapidly reduces sample volume while removing soluble matrix-associated inhibitors- providing more efficient and streamlined laboratory processes. The process is very rapid and allows large sample volumes to be concentrated into a standard analysis volume in minutes.

The concentration process uses microfiltration to capture particles on the surface of a porous membrane filter within the Concentrating Pipette's single-use tip. After the sample has been processed and the particles have been trapped, InnovaPrep's patented Wet Foam Elution™ process is employed to wash the particles off of the membrane surface into a very small liquid volume, to better match the input sample volume required for the chosen rapid detection methods.

EFFICIENCY

The overall efficiency is the percent of target microparticles that are concentrated and eluted (recovered) from the CPT. Several factors can affect efficiency, including the nature of the liquid matrix itself, the shape and nature of the microparticles, charges native to the particles, stickiness of the particles and combinations of other nebulous factors. Efficiency for any type of sample generally has to be determined experimentally, due to these interactions. While efficiency will vary between sample types, efficiency is generally consistent for samples of the same type. Efficiency can also vary based on the analytical method and the variability inherent in either the sample or the analytical method. Efficiencies from 50% to over 90% are common.

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CONCENTRATION FACTOR

The Concentration Factor is the actual mathematical advantage that the Concentrating Pipette provides to you. Concentration factor is calculated
as follows:

1. Calculate the liquid to liquid concentration factor
2. Multiply that figure by the efficiency for the sample that was run.
3. The result is the “CF” or concentration factor for that sample.

TECHNICAL TIP

Note that the higher the initial volume, and the lower the final volume, the higher the CF will be. Efficiency is not the most important factor; it is important, but a high CF is what gives the most target particles to the analytical device, kit, strip, instrument, or whatever analysis method that is used.

Your samples are special. These examples and the following equation will help you determine your options and potential concentration factor.

YOUR SAMPLES

1. ___ mL concentrated to _.__ mL =
___/_.__ = ______X Liquid : Liquid

2. ______X x ___% recovery =
_____ x 0.___ = ______X Concentration Facter (CF)

The highest possible initial sample volume and lowest possible final volume, combined with the best efficiency you can get, will give you the highest concentration factor.

EXAMPLES

Example 1:
1. 100 mL concentrated to 0.25 mL =
100/0.25 = 400X

2. 400X x 80% recovery =
400 x 0.80 = 320X CF

Example 2:
1. 1000 mL concentrated to 0.25 mL =
1000/0.25 = 4000X Liquid : Liquid

2. 4000X x 80% recovery =
4000 x 0.80 = 3200X CF

Processing a larger sample to start with gives a much larger concentration factor.

Example 3:
1. 500 mL concentrated to 0.15 mL = 500/0.15 = 3333X Liquid : Liquid

2. 3333 x 61% recovery =
3333 x 0.61 = 2033X CF

Example 4:
1. 50 mL concentrated to 0.25 mL = 50/0.25 = 200X Liquid : Liquid

2. 200X x 95% recovery =
200 x 0.95 = 190X CF