

# **Centrifugo-pneumatic multi-liquid aliquoting – parallel aliquoting and combination of multiple liquids in centrifugal microfluidics**

## **Supplementary information**

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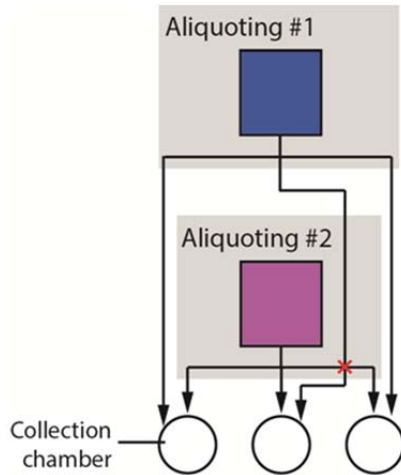
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## Electronic supplementary material

### Geometric limitations

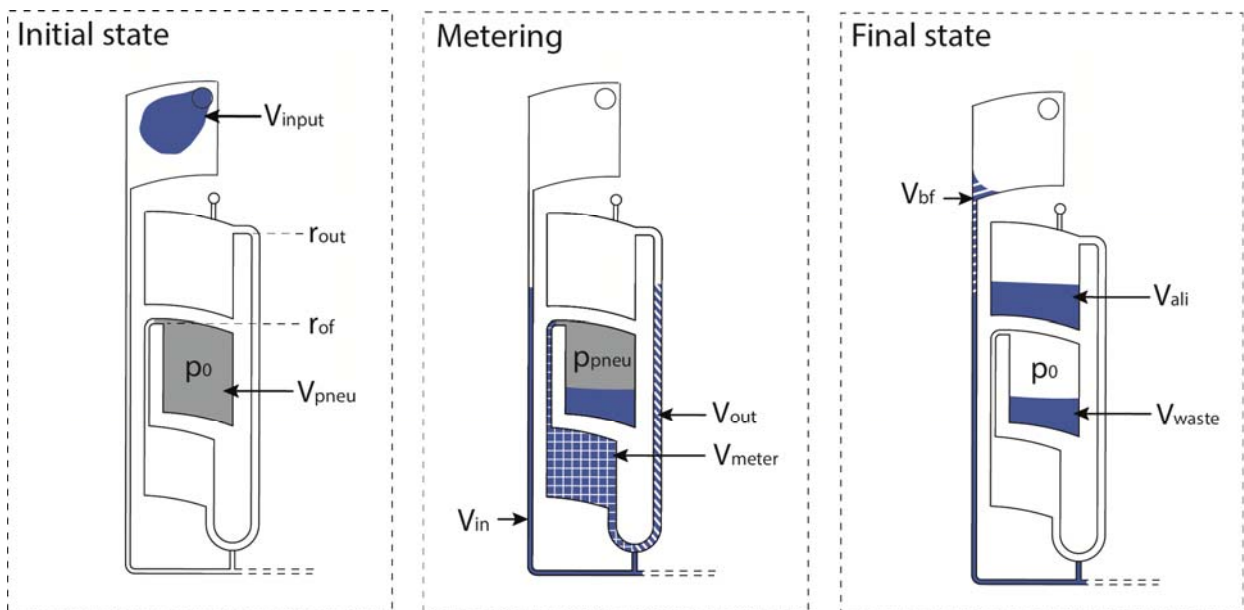
Due to traditional outwards transport in centrifugal microfluidics, pairwise combination of aliquots in more than two collection chambers becomes impossible within a single fluidic layer. ESI Fig 1 illustrates this geometric limitation for two aliquoting structures of three aliquots each.



**ESI Fig 1:** If liquid transport is restricted to outwards transport, pairwise combination in more than two collection chambers from two aliquoting structures becomes impossible within one fluidic layer and without premature contact of the liquids outside of the collection chambers.

### Fluidic design rules

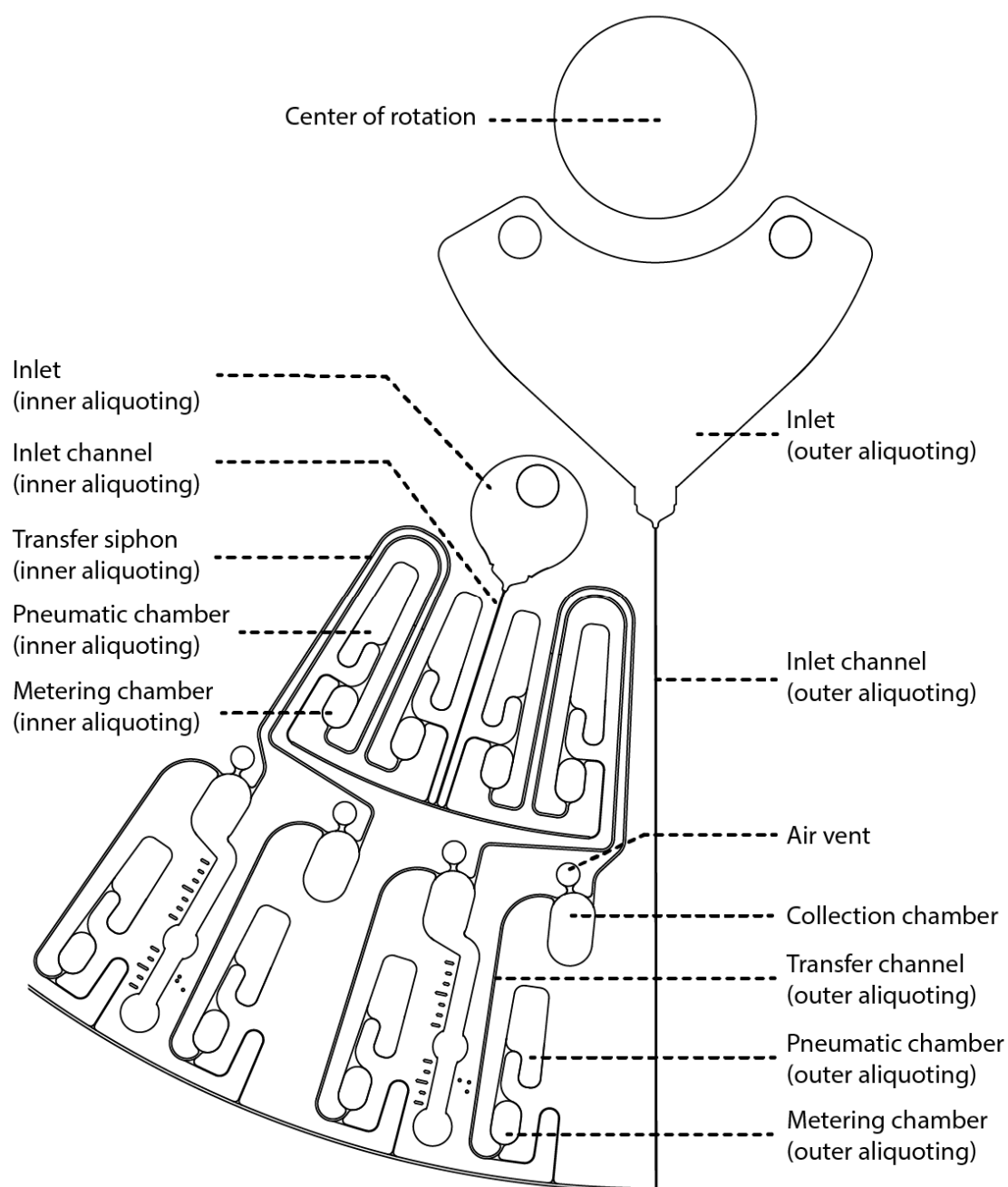
ESI Fig 2 shows the radii and volumes as discussed in the section fluidic design rules of the centrifugo-pneumatic multi-liquid aliquoting.



**ESI Fig 2:** Volumes, pressures and radii as used for derivation of the fluidic design rules.

## Implemented design

ESI Fig. 3 shows an inset of the implemented design including the labels for all channels and chambers.

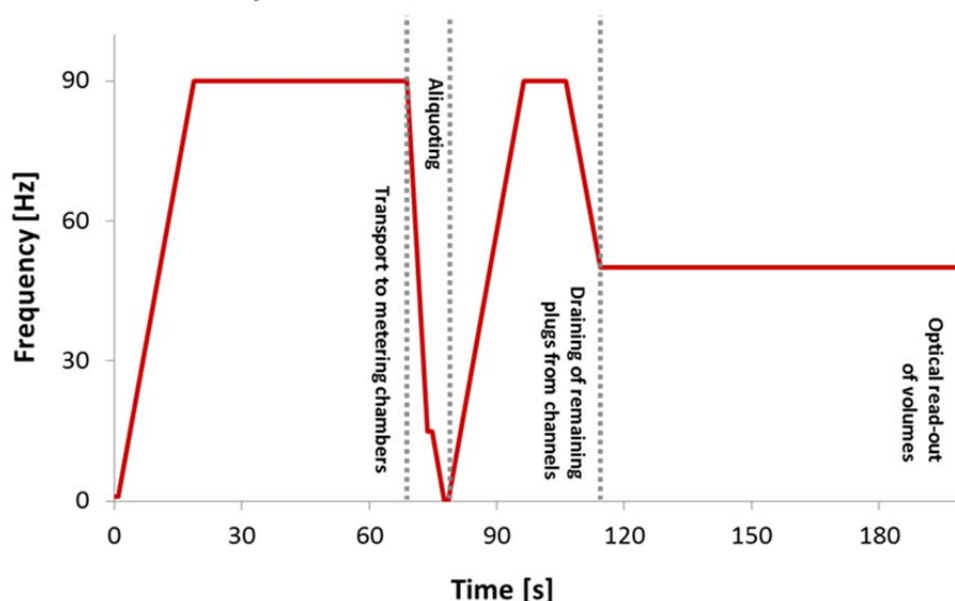


**ESI Fig 3:** *Implemented design including labels.*

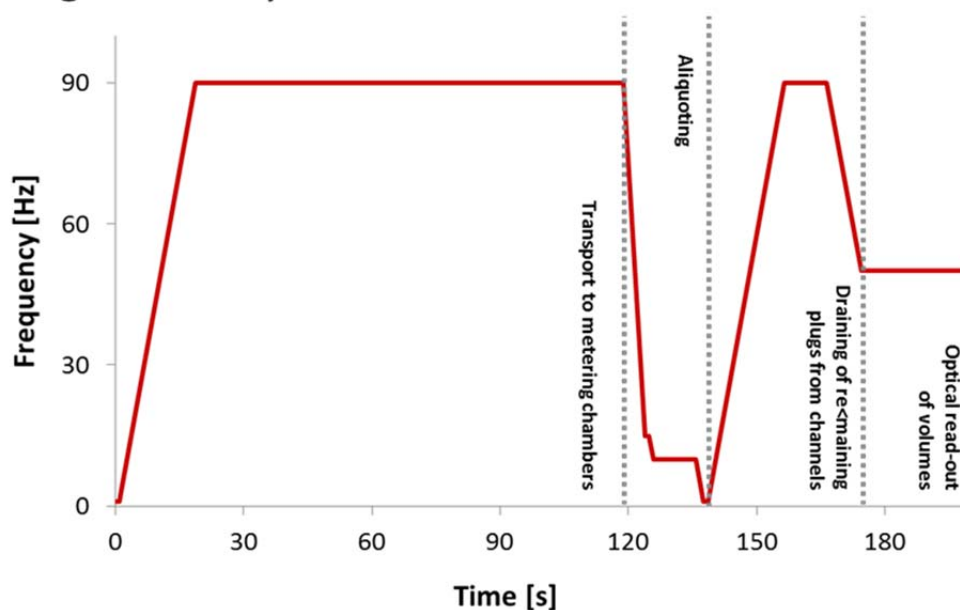
## Frequency protocols

ESI Fig 4 shows the frequency protocols as used in this work. For higher viscosity liquids longer holding times during metering and aliquoting are necessary. The used acceleration is 5 Hz/s. Minimum acceleration is typically not critical, since the high fluidic resistance of the loading channel ensures high pressure loss and little flow during loading. Very low acceleration rates would be critical because then the pressure loss is not sufficient to restrict flow and the liquid level rises above the siphon crest and liquid flows to the collection chambers prior to metering. The minimum required acceleration was determined via network simulations to be 0.25 Hz/s for inner and 0.5 Hz/s for outer aliquoting.

### Low viscosity



### High viscosity



ESI Fig 4: Frequency protocols used for high and low viscosity liquids.

## Calculation of variations

In the following the underlying formulas for the coefficients of variation are given. The parameters M and N give the total number of runs and the total number of aliquots within one run, respectively.

The volume  $V_{m,n}$  is the volume of aliquot n in run m.

Overall CV:

$$CV_{\text{Overall}} = \sqrt{\frac{1}{(M N - 1)} \sum_{m=1}^M \sum_{n=1}^N (V_{m,n} - \bar{V})^2}$$

Inter-run CV:

$$CV_{\text{Inter run}} = \frac{\sqrt{1/(M - 1) \sum_{m=1}^M (\bar{V}_m - \bar{V})^2}}{\bar{V}}$$

Intra-run CV:

$$CV_{\text{Intra run, m}} = \frac{\sqrt{1/(N - 1) \sum_{n=1}^N (V_{m,n} - \bar{V}_m)^2}}{\bar{V}_m}$$

Mean volume of run m:

$$\bar{V}_m = \frac{1}{N} \sum_{n=1}^N V_{m,n}$$

Overall mean volume:

$$\bar{V} = \frac{1}{NM} \sum_{m=1}^M \sum_{n=1}^N V_{m,n}$$