# Investigating cross-linking of sodium alginate and calcium chloride for use in a microfluidic kit.

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## BACKGROUND

- Standardised microcapsules have many potential uses such drug delivery, targeted therapy and diagnostics [1]. Importantly, the materials used in microcapsules are diverse, thus arising creative and novel solutions within a range of industries including the medical, textiles and food fields [2].
- Encapsulating essential oil (EO) cores in semi-permeable shells may provide novel attachment to textiles and prolonged release of the antimicrobial properties they hold giving rise to many healthcare textile uses.
- A novel smart microfluidic control system (*Fluigent, Fr*) will be used to standardise the materials, size (95-160µm) and production rate of the microcapsules produced. However, this system recommends oil surfactant to result in a water-in-oil microbead.

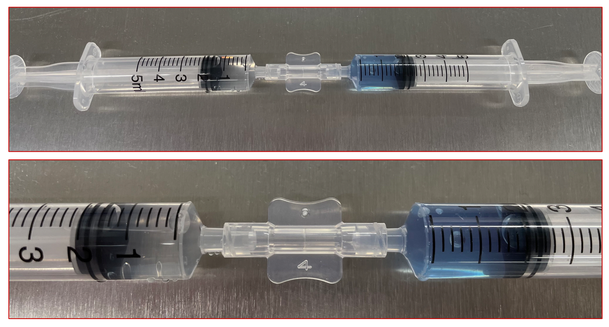


*Figure 1: Luer lock syringe with coupler & solution of SA and $CaCl_2$ each side*

## AIMS & OBJECTIVES

**This project seeks to investigate replacing the disperse surfactant phase with sodium alginate (SA) and a cross-linking agent such as calcium chloride ($CaCl_2$).**

It is paramount that instantaneous polymerisation causing clogging of the microscopic fluidic chamber does not occur, however still producing strong and reliable gels to be used as an encapsulant.

**Parameters and targets considered:**

- Must form a gel but not solidify
- Must take around 1-2 minutes to form - not instant polymerisation
- Must not be too viscous - as has to pass through 250µm diameter tubing

## MATERIALS & METHODS

- SA concentrations (0.5, 1, 1.5, 2%)
- CaCl2 concentrations (5, 10, 15mM)
- 5ml Luer lock syringe + female-female coupler
- Flow EZ ™ (*Fluigent, Fr*)

**Concentrations of sodium alginate and $CaCl_2$ underwent timed tests upon mixing in various contexts:**

1. Extrusion method as seen in [3], using 500ml beaker of $CaCl_2$ and 100µl sodium alginate.
2. Luer lock syringe method: 2ml of each concentrations were added to each syringe and each mix was counted. Gel then underwent inverted vial test modified from [4]
3. Fluigent tubing: The selected concentrations were trialled using the Raydrop™ (*Fluigent, Fr*) microchip.

## RESULTS

- $CaCl_2$ in concentrations >15mM resulted in instant gelation (*figure 2*)
- *Figure 1* also shows how $CaCl_2$ concentrations of 5mM resulted in a prolonged gelation of ~1-2 minutes, however a weak and unstable gel was produced (*figure 4*) which may not be desired for use as a microcapsule shell.
- Concentrations of $CaCl_2$ <5mM were tested against concentrations of SA <1% and which confirmed weak and sub-optimal cross-linking (data not shown).

***Figure 2: The time it takes for crosslinking to occur - method 1***

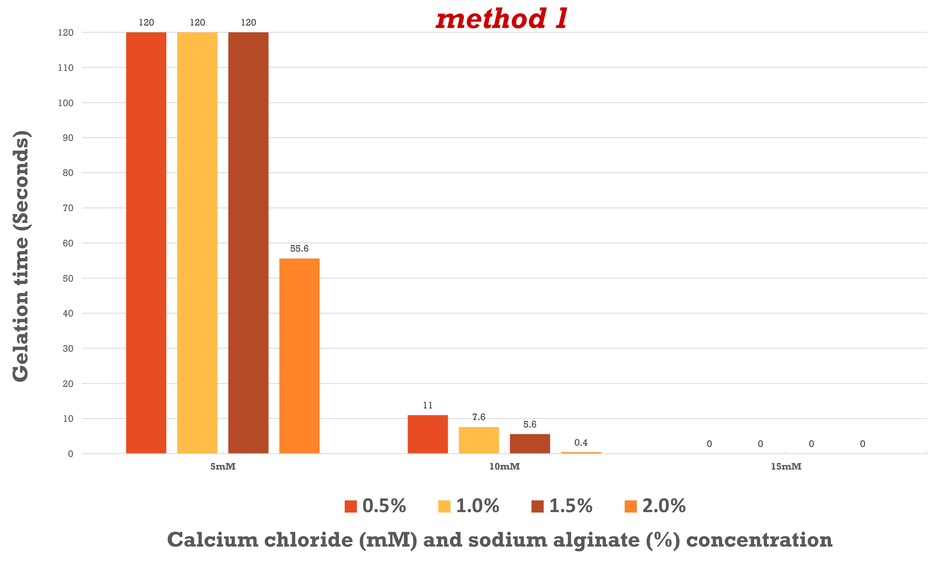
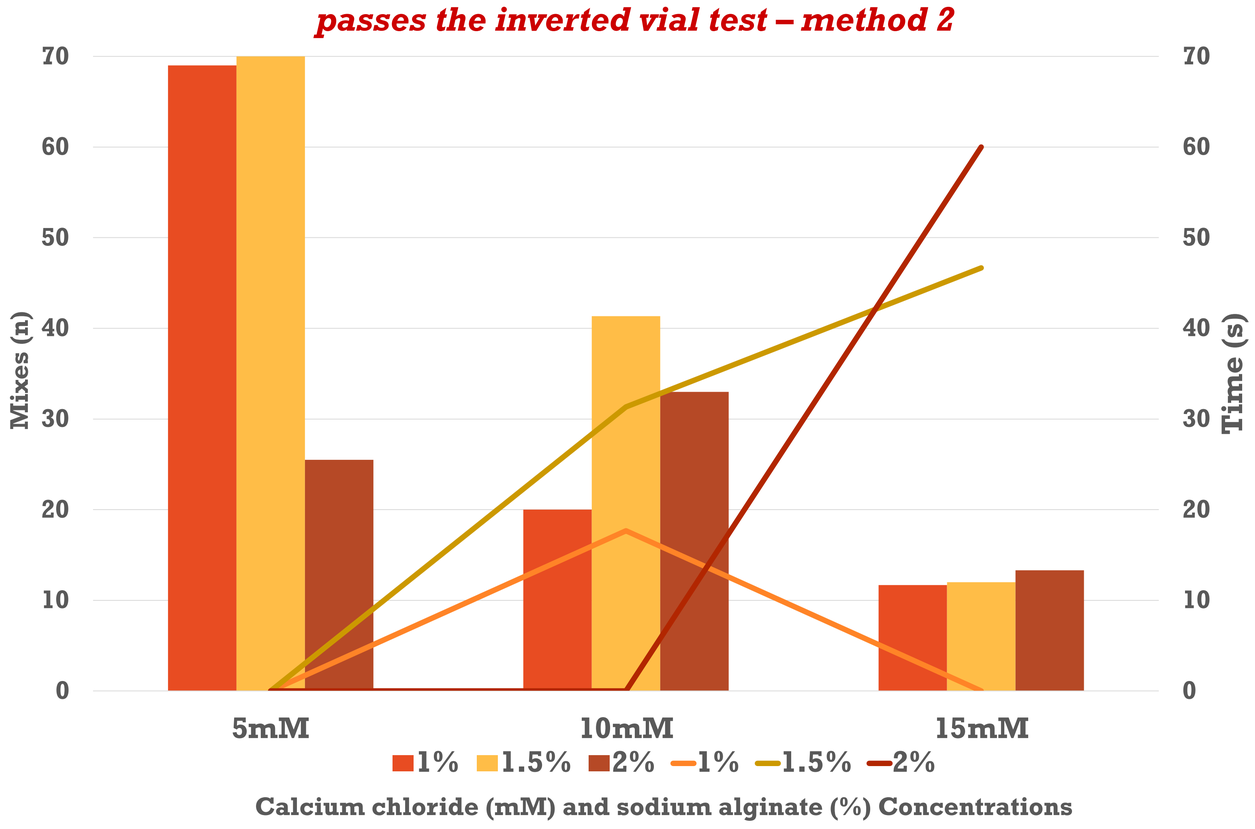



***Figure 3: The amount of 'mixes' it takes to form a gel that passes the inverted vial test – method 2***




## RESULTS

- Figure 3 shows that an increase in time for cross-linking occurs as the concentrations increase, this could be because of higher availability of calcium ions with the alginate polymers.
- Using 2% alginate performs well in the inverted vial test, however upon trialling with the Flow EZ™ tubing requires a pressure over 1bar and takes >2 minutes 30 seconds to be pushed through a length of 60cm (data not shown).

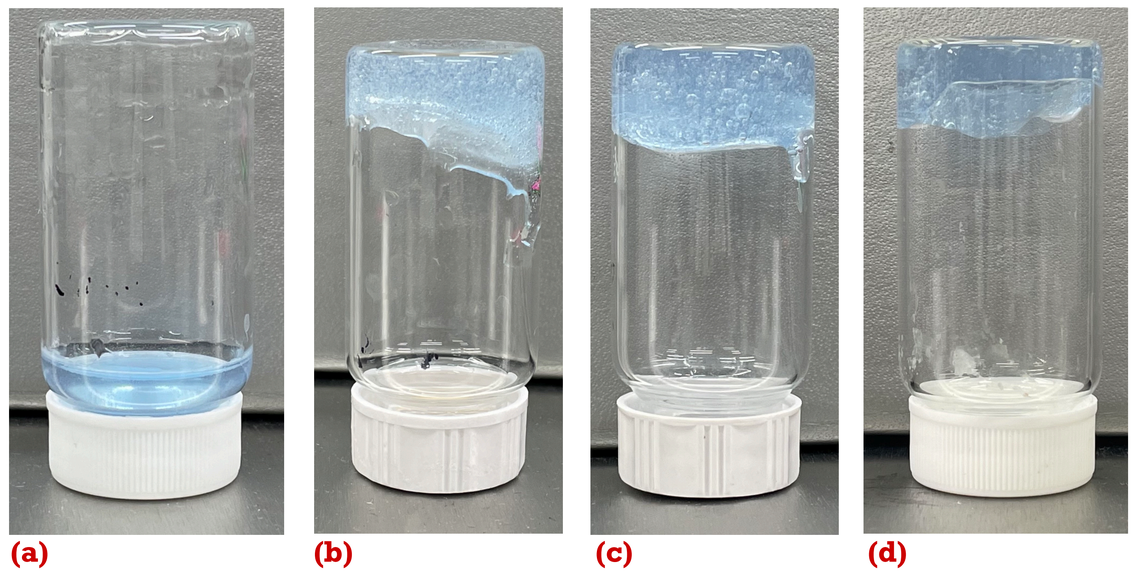



***Figure 4: The inverted vial test (method 2).*** **(a)** $CaCl_2$ 5mM:1.5% SA; **(b)** $CaCl_2$ 10mM:1.5% SA; **(c)** $CaCl_2$ 15mM:1.5% SA; (d) $CaCl_2$15mM:2% SA.

*Figure 4a* shows a liquid gel, as it slid to the bottom of the vial instantly.

According to [4] classification, the gels *b, c* and *d* are polymer gels as they stayed at the top for a considerable amount of time before sliding down the vial at

The gel in *figure 4d* took over 10 minutes to drop to the bottom before ending the experiment.

## CONCLUSION

This study can provide a set of cross-linking timings ranging from instantaneous (0s) to over 2 minutes, making it possible to use in a microfluidic kit without setting and blocking the system.
There is a possible hypothesis for using calcium chloride in the dispersed phase of the Flow EZ (Fluigent, Fr), to improve cost and ease when using the kit and this will be investigated further.

## REFERENCES

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[4] Sharma, P.K., Reilly, M.J., Bhatia, S.K., Sakhitab, N., Archambault, J.D. and Bhatia, S.R., 2008. Effect of pharmaceuticals on thermoreversible gelation of PEO–PPO–PEO copolymers. *Colloids and Surfaces B: Biointerfaces*, *63*(2), pp.229-235.