Surface modification of sustainably-formulated bacterial cellulose nanoparticles for drug delivery

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Background

- We developed bacterial cellulose nanoparticles (**BCNPs)** for sustainable drug delivery, motivated by cellulose's **bio-renewable sourcing** and **environmentally friendly end-of-life**, and **biocompatibility**.[1, 6]
- Surface modified BCNPs provide a small library to incorporate a range of drugs.
- Tuning the nanoparticle surface provides control over the nanoparticle's chemical & physical properties which can influence interactions with drugs, cells, and tissue.
- Our objective in this study was to modify BCNPs with the surface functional groups: acetyl-, methyl-, and amin- (Figure 1).

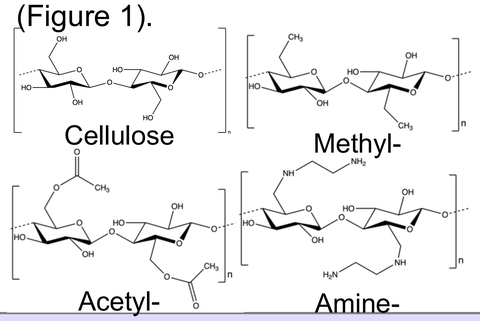


Figure 1. BC surface modification reactions include methylation (methyl-),[4] acetylation (acetyl-),[2] and amination (amin-)[3].

Methods: nanoparticle synthesis

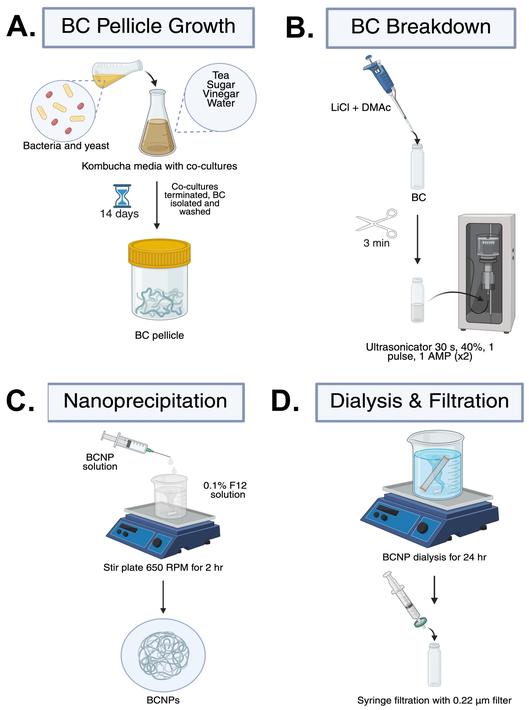


Figure 2.
(A) Grow bacterial cellulose (BC) pellicle in kombucha for 14-days then isolate and wash.
(B) Dissolve BC chemically & mechanically.
(C) Formulate BC nanoparticles (BCNPs) via nanoprecipitation and surfactant solution.
(D) Dialyze BCNPs and then syringe filter for size.

Methods: *ex vivo* model preparation

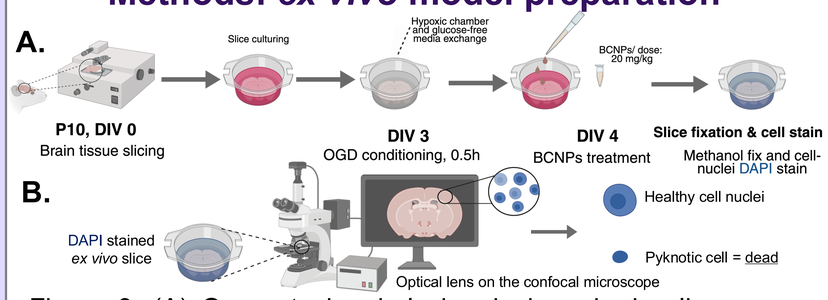


Figure 3. (A) Organotypic whole hemisphere brain slices were prepared and cultured for 4-days *in vitro* prior to oxygen-glucose deprivation (OGD) and applying BCNPs. (B)Slices were then imaged on a confocal microscope at 60x and 240x.

Nanoparticle physical-chemical properties

Table 1. BCNPs mean particle size (nm) and surface charge (mV) with the standard error of the mean (SEM).

Sample	Mean particle size ± SEM (nm)	Zeta-potential ± SEM (mV)
BCNPs	114.0 ± 7.2	-25.9 ± 2.6
Acetyl-BCNPs	117.3 ± 6.7	-19.4 ± 6.5
Methyl-BCNPs	102.1 ± 1.5	-16.0 ± 2.0
Amin-BCNPs	107.9 ± 3.8	-19.9 ± 2.8

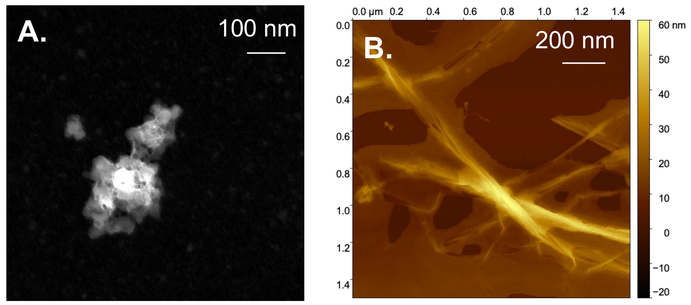


Figure 4. (A) Scanning transmission electron microscopy of opt-BCNPs, scale bar set to 100 nm & (B) atomic force microscopy of BC fibrils that form nanoparticles, scale bar set to 0.2 µm.

Conclusion

We demonstrated that we can formulate sub-120nm BCNPs with slight **negative surface charge** for targeted drug delivery. We show that BCNPs can encapsulate a hydrophobic drug with **high EE.** Lastly, we demonstrated that BCNPs **localized in cells** of therapeutic interest (microglia) in cultured brain slices. Future work includes assessing the drug release profile *in vitro* and evaluating therapeutic efficacy in our *ex vivo* model.

Drug loading (DL) and encapsulation efficiency (EE)

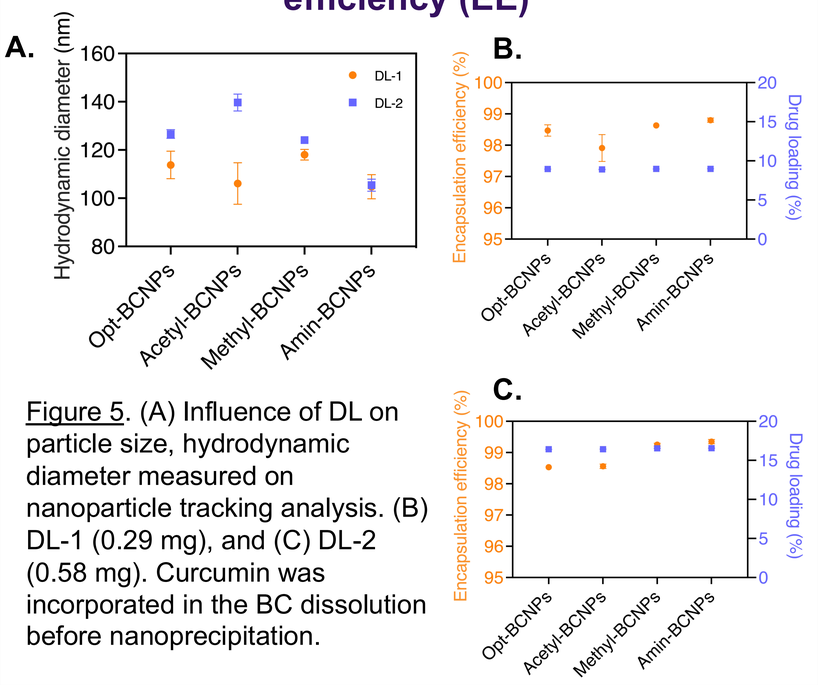
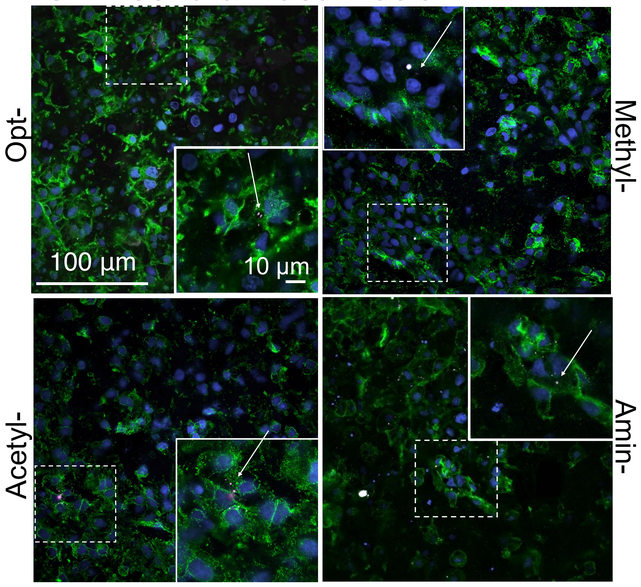


Figure 5. (A) Influence of DL on particle size, hydrodynamic diameter measured on nanoparticle tracking analysis. (B) DL-1 (0.29 mg), and (C) DL-2 (0.58 mg). Curcumin was incorporated in the BC dissolution before nanoprecipitation.

BCNP cellular localization

Figure 6. BCNPs labeled with Carbotrace localized and associated with iba-1+ cells in the cortex region of the brain tissue. Cell nuclei (DAPI).



References: [1]**Balistreri** et al, RSC, 2024, [2]Chen et al, ACS, 2019, [3]Ni et al, Chemical Engineering Journal, 2023, [4]Vierira, Brazilian symposium, 2004, [6]Balistreri, et al., *manuscript in preparation*. **Acknowledgments**: Ian Campbell & Nels Schimek for Python training, Julia Amorim and Jennifer Tran for kombucha & co-culture, and Brendan Butler for *ex vivo* model training. **Funding**: This work was supported by an NSF GRFP (G. Balistreri) and Bindra Endowed Development Professorship.