

Arrest in Coarsening in Biological Liquid-Liquid Phase Separation – or – ‘It ain’t necessarily so ...’

1. **Hydrodynamically Interrupted Droplet Growth in Scalar Active Matter**

Authors: Rajesh Singh and M. E. Cates
Phys. Rev. Lett. **123**, 148005 (2019)

2. **Physics of Active Emulsions**

Authors: Christoph A Weber, David Zwicker, Frank Jülicher, and Chiu Fan Lee
Rep. Prog. Phys. **82**, 064601 (2019)

Recommended with a Commentary by Tom McLeish, Department of Physics, University of York, UK

There is a rather serious collective phenomenon going on at the moment in the physics of liquid-liquid phase separation (LLPS) in the form of a widespread excitement about its application to cell biology. For as well as membrane-bound organelles encapsulating subsystems with specific functions within the cell, there is growing recognition of a wide class of ‘membrane-free organelles’ whose specific liquid content has spontaneously separated and self-assembled within the intracellular medium. Examples are known as ‘stress-granules’, ‘P-bodies’, ‘aggresomes’, ‘replication clusters’. They are implicated in many processes, including protein manufacture, photosynthesis, sequestration and enzyme function. Careful *in vivo* and *in vitro* experimental programmes have indicated that in many cases at least, thermodynamic phase separation of some kind is implicated. Photobleaching and other techniques has verified the liquid nature of the phase-separated regions. Their morphology is typically ‘nucleated’, rather than canonically ‘spinodal’, which is a significant observation yet rarely commented upon. For a helpful review of biological LLPS, see [1].

There are many special concerns and questions that arise in these biological cases of LLPS, even if this proves to be the correct overall physical framework in which to understand such ‘membrane-less organelles’. One in particular, that has appeared in many of the relevant publications recently, is the necessity that the canonical coarsening (or ‘Ostwald ripening’) of phase-separated regions be arrested early in the context of biological cells. The various functions of the membrane-free organelles typically require that their droplet morphologies do not grow without bound, and in fact remain significantly smaller than the size of a single cell. The problem is that each approach to this problem has tended to encounter one mechanism, naturally associated with that approach by which finite-size LLPS arrest might be achieved, which might be interpreted as typical. There has not yet been a chance to compare and evaluate competing mechanisms in a coordinated way (of course, it is perfectly possible, and even likely, that different biological systems have recruited different physics to achieve the necessary arrest of coarsening and final morphology). Both invoke the possibility afforded by living, but not inert,

systems of being local driven. The formulations then may break detailed balance, and time-reversal symmetry, without necessarily displaying explicit models of the molecular agents of driving (e.g. enzymes or molecular motors, or on an entirely different scale, microscopic ‘swimmers’). Here I make a step towards the comprehensive review that we need, by contrasting two recent theoretical approaches.

Singh and Cates in paper 1 take a phenomenological route to an active version of canonical phase separation, asking what (non-thermodynamic) terms may be added (as usual in order of powers of the field variable) in Hohenberg’s and Halperin’s ‘model-H’ [2] for the thermodynamic behavior of two-component liquid systems with conserved order-parameter. The starting point is the usual near-equilibrium expression for the current of the conserved field $\phi(\mathbf{r})$ under a mobility M :

$$\mathbf{J}(\mathbf{r}) = -M\nabla\mu(\mathbf{r}) + \sqrt{2MD}\Lambda \quad ; \quad \mu = \frac{\delta\mathcal{F}}{\delta\phi} \quad (1)$$

where μ is the chemical potential field, given by the functional derivative of a free-energy density in the normal model-H. Λ is an uncorrelated Gaussian field of white noise of effective temperature D . The authors add the simplest new terms in \mathbf{J} and μ that break time-reversal symmetry in (1) to create their ‘active model-H’. They take the form

$$\mathbf{J}_\zeta(\mathbf{r}) = M\zeta(\nabla^2\phi)\nabla\phi \quad ; \quad \mu_\lambda = \lambda|\nabla\phi|^2 \quad (2)$$

The authors analyse the rich phenomenology of this active model-H in terms of the two extra ‘active tensions’ that it gives rise to in the nonlinear regime of droplet growth and stabilization. They find that the new ‘active’ physics offers several mechanisms for arrest, or effective arrest, of coarsening, or equivalently the emergence of a preferred length scale:

- (i) A ‘self-shearing’ instability, driven by activity at the interfaces stretching them, drives droplets to self-breakup. The preferred size is found at the balance point of diffusive growth and breakup.
- (ii) A ‘reverse Ostwald ripening’ regime in which interfaces are driven to a preferred local curvature by negative values of one of the driven effective surface tensions. This was previously noticed in this approach to an ‘active model-B’ [3]
- (iii) A combination of these effects which leads to uniform and small droplets.

A fourth regime simply renormalizes passive phase separation. Figure 1 (reproduced from paper 1) illustrates the morphologies that arise from the dominance of these processes at different values of the effective tensions.

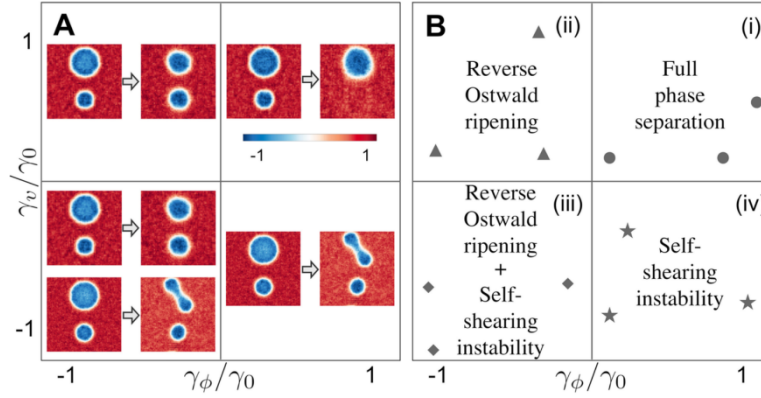


Figure 1: Phases of active model H of paper 1 in the plane of the two effective active tensions. Panel A shows the dynamics of two proximal and unequal sized bubbles in the four regimes indicated in panel B.

Paper 2 by Weber *et al.* is an example of another direction of attack on the problem of living, or active, phase separation. In place of starting from a thermodynamic field theory and asking what simplest phenomenological terms within that formalism would represent active, dissipative content, these authors suppose a particular structural dynamics. The relevant section of this paper is section 4 on ‘chemical reactions’ – until that point it covers, as its title makes clear, a great deal of material in review. The effective role of such reactions is to provide an alternative route to increasing the concentration of component A relative to B in addition to convection by the current \mathbf{J} of eqn. (1) – it may interconvert from A to B locally by a reaction. Such interconversion may be thermodynamically ‘downhill’ through by a chemical potential and a reaction rate, or driven by active consumption of an implicit molecular fuel. In this case, the key structure is best seen by its modification to the dynamical equation of the composition field ϕ :

$$\frac{\partial \phi(\mathbf{r}, t)}{\partial t} + v \cdot \nabla \phi = -\nabla \cdot \mathbf{J}(\mathbf{r}, t) + s(\phi) \quad (3)$$

Now the activity is inserted into the local term. It is possible to break detailed balance in different ways, which the authors term ‘external maintenance’ and ‘internal maintenance’ signifying the positive creation of droplet material external to the droplet (feeding it though inward flux) or internally to it. The mathematical structure of these two alternatives in linear response looks like:

$$s(\phi) = \begin{cases} \Gamma_{in} - k_{in}(\phi - \phi_{in}^0) & \text{inside} \\ \Gamma_{out} - k_{out}(\phi - \phi_{out}^0) & \text{outside} \end{cases} \quad (4)$$

Here the bracketed terms represent perturbations of the field from its equilibrium values inside and outside the droplet, and the relaxation rates of those differences respectively. The Γ -terms are generative ‘basal fluxes’ that create or annihilate the field, requiring driven, dissipative processes to do so. ‘External maintenance’ refers to the case in which $\Gamma_{out} > 0$, $\Gamma_{in} < 0$ (vice versa

for the other case). The interesting observation is that arrest of Ostwald ripening and the emergence of a stable droplet size arises in the case of external, but not internal, maintenance (in the latter case droplet size diverges either side of an unstable fixed point). In cases of both single and multiple drops, the stability of droplet size to a radius R_{stab} originates from a competition of the destabilising effect of surface tension, which tends to increase R_{stab} , and the stabilising effects induced by the diffusive fluxes driven by the chemical reactions. Note that this describes a different underlying physics to the stable droplet phase of paper 1, in which the surface tension picks up a (radius dependent) negative term.

Interestingly, in spite of the difference in underlying physics, the similarities of the two models in their emergent phenomenology extends to a regime of driven droplet breakup (see Figure 2). In this case, the breakup instability appears at sufficiently high supersaturation in the solvent, and arises from a local coupling of droplet curvature and inward flux (positive and destabilising in the case of external-maintenance). While similar to regime (a) of paper 1, it does not seem possible to map this reaction-diffusion system onto the dissipative phenomenology of active model-B.

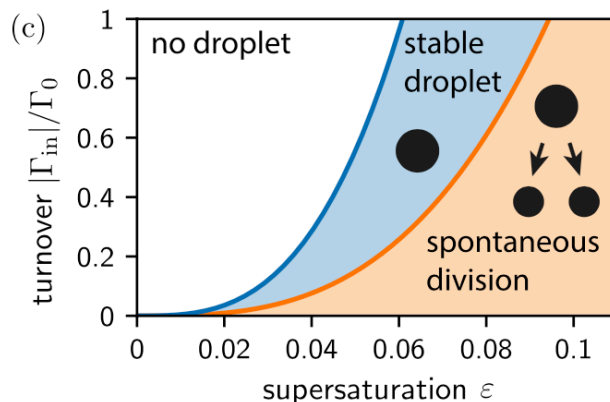


Figure 2: Phases of the active reaction model of paper-2 (from figure 14 of Weber *et al.*)

The conclusion of this brief comparison is that similar emergent properties of active LLPS systems does not imply equivalence of underlying physics. In fact, there are many possible routes to arrest of coarsening and selected droplet size, which does not even require activity (*e.g.* through a balance of long- and short-range potentials [4]).

References

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- [4] Sweatman M.B. and Lue L. 'The giant SALR cluster fluid: a review', *Adv. Theo. Sim.*, 900025 (2019)