

CLAIMS

What is claimed is:

1. A method of identifying a compound that modulates a characteristic associated with one or more condensates comprising a RBM20 polypeptide (“RBM20 condensates”) in the cytoplasm of a cell, the method comprising:
 - (a) admixing the compound and a composition comprising the cell,
wherein (i) the cell comprises the one or more RBM20 condensates, and/or (ii) the one or more RBM20 condensates form in the cell after the compound contacts the composition;
 - (b) direct or indirect labeling of the RBM20 polypeptide;
 - (c) imaging at least a portion of the composition comprising the cell,
wherein detected light is due to direct labeling or indirect labeling of the RBM20 polypeptide, and
wherein the imaging provides data to assess the characteristic associated with the one or more RBM20 condensates; and
 - (d) determining the characteristic associated with the one or more RBM20 condensates in the cytoplasm using the imaging,wherein a modulation in the characteristic, as compared to a reference, indicates that the compound modulates the characteristic associated with the one or more RBM20 condensates in the cytoplasm.
2. The method of claim 1, wherein the characteristic associated with the one or more RBM20 condensates is based on any one or more of the following:
 - (i) location of the one or more RBM20 condensates;
 - (ii) distribution of the one or more RBM20 condensates and/or the RBM20 polypeptide;
 - (iii) number of the one or more RBM20 condensates;
 - (iv) size of the one or more RBM20 condensates;
 - (v) ratio of the amount of one or more RBM20 condensates and a reference condensate;
 - (vi) a functional activity associated with the one or more RBM20 condensates;

- (vii) composition of the one or more RBM20 condensates;
 - (viii) co-localization of the one or more RBM20 condensates with a biomolecule;
 - (ix) diffusion coefficient of a component of the one or more RBM20 condensates;
 - (x) stability of the one or more RBM20 condensates;
 - (xi) dissolution or reduction in size of the one or more RBM20 condensates;
 - (xii) surface area of the one or more RBM20 condensates;
 - (xiii) sphericity of the one or more RBM20 condensates;
 - (xiv) liquidity of the one or more RBM20 condensates; and
 - (xv) solidification of the one or more RBM20 condensates.
3. The method of claim 2, wherein the modulation in the characteristic is based on a decrease in the number of the one or more RBM20 condensates in the cytoplasm of the cell.
 4. The method of claim 2 or 3, wherein the modulation in the characteristic is based on dissolution or reduction in size of the one or more RBM20 condensates in the cytoplasm of the cell.
 5. The method of claim 1, wherein the characteristic associated with the one or more RBM20 condensates comprises location of the one or more RBM20 condensates, distribution of the one or more RBM20 condensates, number of the one or more RBM20 condensates, size of the one or more RBM20 condensates, and ratio of the amount of one or more RBM20 condensates and a reference condensate.
 6. The method of any one of claims 1-5, wherein the RBM20 polypeptide is a wild type RBM20 polypeptide.
 7. The method of any one of claims 1-5, wherein the RBM20 polypeptide is a mutant RBM20 polypeptide.
 8. The method of claim 7, wherein the mutant RBM20 polypeptide comprises a mutation in an intrinsically disorder region (IDR) or an RS-rich region.

9. The method of claim 7 or 8, wherein the mutant RBM20 polypeptide comprises a mutation in one or more of the following positions: arginine 634, serine 635, arginine 636, serine 637, and proline 638, wherein the positions are relative to SEQ ID NO: 1.
10. The method of claim 9, wherein the mutant RBM20 polypeptide comprises one or more of the following mutations: R636S, R636C, R636H, R634Q, S637G, P638L, S635A, S635E, and S637E, wherein the positions are relative to SEQ ID NO: 1.
11. The method of claim 7, wherein the mutant RBM20 polypeptide comprises one or more of the following mutations: E913K, R716Q, and V535L, wherein the positions are relative to SEQ ID NO: 1.
12. The method of any one of claims 1-11, wherein the RBM20 polypeptide is heterologously expressed in the cell.
13. The method of any one of claims 1-11, wherein the RBM20 polypeptide is homologously expressed in the cell.
14. The method of any one of claims 1-13, wherein the cell is a model of a cardiac cell type.
15. The method of any one of claims 1-13, wherein the cell is selected from the group consisting of: a HeLa cell, a U2OS cell, a human embryonic kidney cell, a human induced pluripotent stem cell, and a stem cell.
16. The method of any one of claims 1-15, wherein the cell is homozygous for alleles encoding the RBM20 polypeptide.
17. The method of any one of claims 1-15, wherein the cell is heterozygous for alleles encoding the RBM20 polypeptide.
18. The method of any one of claims 1-17, wherein the reference comprises an aliquot of the composition comprising the cell admixed with a negative or positive control reference compound.
19. The method of any one of claims 1-18, wherein the method further comprises determining one or more cellular features of the cell.

20. The method of any one of claims 1-19, further comprising contacting at least a portion of the composition or the cell with a fixative.
21. The method of any one of claims 1-20, further comprising contacting at least a portion of the composition or the cell with a stain.
22. A method of identifying a compound useful for treating a RBM20-associated disease, the method comprising identifying a compound according to any one of the methods of claims 1-21.
23. The method of claim 2, wherein the modulation in the characteristic is based on the number of the one or more RBM20 condensates or the dissolution of the one or more RBM20 condensates.
24. The method of claim 2, wherein the modulation in the characteristic is based on the number of the one or more RBM20 condensates and the dissolution of the one or more RBM20 condensates.