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Some More Gap Areas of Investigation on Exploring Affect of Invisible Force of Self Gravity in Living Mass

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Illustration Table 1.

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Abstract

On the principle of abductive reasoning through successive approximation on sporadic set of evidences; explanation is sought on the possible role of invisible force of self gravity in biological mass. Self organization of biological macromolecules is affected by self gravity. Nucleic acid having highest molar mass is naturally attracted on priority among other macromolecules and is compressed to the centre of the cell by the force of self gravity making it tightly packed under free floating fluidic cellular environment. Proteins having intermediate molecular weight preferably remain in-between position of the cell. Fats and lipids having lowest density would stay behind, especially at the periphery in the cell membrane under principle of 'less dense material - lesser attraction of self gravity'. Protein sphericity is assumed when the strength of dipole moment gets weaker with increase in distance from central position, tensile and compressive gravity gradients of self gravity might get increase with increase in mass and tend to fold the backbone of the polypeptide chains to a globular conformation at the 'farthest point' similar to 'fountain effect' with 'central tendency'. Native conformation, denaturation and renaturation of protein are thought to be due to self gravity for getting united or reunited after withdrawal of external force that would cause denaturation of protein. Elongated fibrous proteins can also attain sphericity when local effect can be augmented. Equation of state under hydrostatic equilibrium can explain phenomena like heavier cells to grow faster than lighter cells or cell density to increase prior to bud formation.

Introduction

On the basis of interaction after publication of the article[1] 'Invisible force of self gravity: A gap area of investigation in life science', it has been observed that researchers, especially working at macromolecular level, are yet to comprehend the concept of self gravitation bio. We continue on the principle of abductive reasoning through successive approximation on sporadic set of evidences to explore invisible force of self gravity in biological mass, especially at macromolecular level, as gap areas of investigation. Before going through the present article, one however may go through the previous article, as above, for getting comprensive idea on self gravity and its role on living mass.

Self gravity dictates self organization of macromolecules in living cell

Macromolecules are important for various biological functions. There are four basic kinds of biological macromolecules. These are carbohydrates, lipids, proteins and nucleic acids. These polymers are composed of different monomers. But one thing might get less attention that these macromolecules are having different molar mass or molecular weight in addition to density and under free floating condition, macromolecules having higher molar mass or higher density will occupy the core position of self gravitating body and macromolecules having intermediate or lesser molecular weight or lesser density will remain away from core. That is 'higher the density- higher would be the attractive force of self gravity' or in reverse way 'lesser the density - lesser would be the attraction of self gravity'. This is what is happening in the movement or position of macromolecules in the living cells in general, which testify that self gravity is operating in the living cell without any doubt. Alternative justifications in this regard are either absent or not elegant and well designed.

Let us assume that release of water molecules and presence of salts and other materials are ensured in the fluids from carbohydrates, proteins and other sources. Buoyant like forces automatically come into operation as a consequence of accumulation of fluid to a particular depth, thereby separating the rest biomass from the inertial external gravitational force. Under lifted or free floating condition, biological macromolecules having higher molar mass and density would be attracted first at the self gravity's core, as higher the mass, higher would be the

gravitational attraction. Compared to nucleic acid, molar mass of carbohydrate is less. For instance, molar mass of say, galactose $C_6H_{12}O_6$ is 180 g/mol. The molar mass, density and solubility of another carbohydrate $C_{12}H_{22}O_{11}$ is 342.297 g/mol, 1.723 g/cm^3 and 683.0 g/L respectively. On the other hand, molar mass of water (H_2O) is 18 g/mol, sodium chloride (NaCl) - 58.443 g/mol and so on. Density of water is 1g/ml at 4^{0}C. So carbohydrates, water, salts possibly play effective primary role as 'metabolically inert infrastructure' or as foundation over which other macromolecules can ballet as per dictation of self gravity.

In case of nucleic acid, molar mass of DNA fragments etc. is highest say, 1000–5,000,000 g/mol. Therefore under near free floating condition, nucleic acid which has higher molar mass or molecular weight as well as density, would tend to remain near the central core due to attraction of self gravity. It is to be noted that nucleic acids contain phosphorus, in addition to C, H, N & O. Unlike proteins, nucleic acids contained no sulfur. The DNA polymer is much larger and may extend up to 2 meters in length. The nucleus is only about 5μm in diameter. The chromosomal DNA is packed tightly and fit in that small volume. The molecular weight of double stranded DNA is approximately 660 x the number of base pairs. The genome of *E. coli*, for instance, contains 4,639,221 base pairs. The molecular weight of one *E. coli* genome, therefore, would be 660 x 4,639,221 = 2,840,000,000 g/mole. Molecular weights for the DNA from multicellular organisms are commonly 10^9 or greater. The DNA from the smallest human chromosome is over ten times larger than *E. coli* DNA. Therefore accumulation of nucleic acid under tightly packed condition at near central position possibly demonstrates presence of invisible force of self gravity without any contradiction.

Proteins which have intermediate molar mass or molecular weight as well as being comparatively less denser than nucleic acid, would remain in intermediate position under free floating condition as well as under self gravitating environment. In case of fats and lipids, molar mass are also intermediate. But the density of fats and lipids is less than protein on equal volume basis. So fats and lipids are under duress of self gravity to occupy the peripheral position under 'free movement' (free fall) condition. "Lesser the density - lesser would be the attarction of self gravity". Hence finding of lipids in cell membrane is not accidental, but a simple instance of comparatively delayed action of self gravity in attracting less dense materials compared to high dense materials at particular point of time (Illustration 1).

Generally centrifugation is an inverse or opposite process of central attraction of gravitation on any mass. Differential centrifugation involving multiple centrifugation steps is generally used to separate cellular materials (known as homogenate in proteomics). Separation is based on size and density against a saturated sucrose pad. Nuclei and mitochondria (having nucleic acid) will sequentially pellet (precipitate) at low speeds and short intervals such as velocities of 1,000 times the gravitational force on the surface of the earth for 5 minutes. For determination of molecular weight of protein, ultracentrifugation; i.e., spinning in a centrifuge at velocities up to about 60,000 revolutions per minute with centrifugal forces of more than 200,000 times the gravitational force on the surface of the earth is needed. Smaller cell fragments and organelles remain in the supernatant and require more force and greater times to pellet. Presence of two gravitational entities can therefore, be visualized in the process. Pre-centrifugation materials are arranged as per density gradient influenced by the major gravitation field of self gravity of the cell. Post-centrifugation order of succession possibly depicts in vivo sedimentation or natural setting as per earth's gravitational fields. Hence biologists may simply rectify their viewing angle to establish and formally recognize self organization of macromolecules in a cell as due to self gravity, as experimentally as well as in practice; it is a well established fact beyond doubt.

The molecular weight or molar mass and density of some amino acid and fatty acid are shown in illustration as Table 1 for ready reference.

Globular protein form and self gravity

It is interesting that protein sphericity is not yet well defined though many studies are being conducted. Globular shapes, which are close to a sphere, often called spheroproteins, act as enzymes, hormones, transporters of other molecules, stocks of amino acids, and other roles, and they are the most interesting proteins in the design of drugs and understanding of life phenomenon. Let us extend the idea of self gravity towards such globular architecture of protein.

Average protein density is a molecular-weight-dependent function[2]. The spatial average density of proteins can be considered equal to 1.35 g/cm^3 independent of the nature of the protein and particularly independent of its molecular weight. It

is worthy to mention that proteins are composed of hydrophobic and hydrophilic amino acids. As far as molar mass and density are concerned, there is no remarkable difference between hydrophobic and hydrophilic amino acids (illustration in Table 1). Hydrogen bonding between different atoms provides required force. However under free floating native aqueous environment, hydrophobic amino acids get buried in the core of a protein as 'communal aggregation' forming bonding between them, whereas hydrophilic amino acids could remain in the boundary of a protein for interaction with the aqueous solvent forming spherical structure. Thus being shielded by hydrophilic amino acids in the aqueous solvent, the hydrophobic amino acid seems play a crucial role of sphericity with packing interactions on forming 'communal aggregation'[3]. This general scenario sometimes gets changed. For example, the protein myoglobin[4] contains 0.34 gram of iron in 100 grams of protein. The atomic weight of iron is 56; thus the minimum molecular weight of myoglobin is (56 × 100)/0.34 = about 16,500. The minimum molecular weight of hemoglobin that contains four atoms of iron is 4 × 16,500 or 66,000. Thus if a protein contains only one molecule of one of the amino acids or one atom of iron, copper, or another element, the minimum molecular weight of the protein or a subunit differs and thereby their behavior in local self gravitating environment may slightly differ.

Native conformation, denaturation and renaturation of protein vis-a-vis self gravity

After proteins get stabilized by hydrogen bonds, as revealed from above, the strength of dipole moment gets weaker with increase in distance from the central position. At that state, tensile and compressive gravity gradients of self gravity possibly gets increase with increase in mass. Therefore gravity gradients in the backbone of the polypeptide chains possibly tend to fold to a globular conformation at the 'farthest point' similar to 'fountain effect' with 'central tendency', thereby leading to folding in 3-dimensional secondary, tertiary or even quaternary structures of native conformation. However comparative increase and decrease in strength in self gravity and dipole moment at perticular period of time is a matter of investigation. But cirumstatial evidences as narrated below favour the aforesaid conjecture.

It is worthwhile to note that formation of native conformation of protein cannot be a chemical process, but seems to be a physical process, as can be seen from the 'denaturation' and 'renaturation' phenomena of proteins. The reverse process of native state in secondary and tertiary structure of protein is called as 'denaturation'[5]. For protein 'denaturation', there is a need for application of some external physical stress or making protein thermally unstable possibly to the extent of overcoming gravity barrier of the self gravitating mass or putting in a compound such as a strong acid or base, a concentrated inorganic salt, an organic solvent like alcohol or chloroform. Strong acid or base or similar chemical reactions can be considered as short term explosion that can overcome barrier of self gravity. It is important to note that on short term basis, gravity is weaker than electrostatic force. For instance, at electron level, the gravitational force between two electrons is 42 orders of magnitude (10^{42}) weaker than their electrical repulsion. Though these studies are at electron level, but at macromolecular level, report of such comparative study is scanty. It is to be remembered that most of the materials at macromolecular level, that are involved in the biological process, are composed of equal amount of positive and negative electric charges whose forces cancel each other out. Whereas electric and magnetic forces are clearly bipolar, gravity is generally assumed to be always attractive so that no analogous cancellations occur. Therefore gravity works on mass at macromolecular level without any time frame. It is clear that protein aggregation is a mass based aggregation (not charge based). Folding starts after formation of some critical mass that spread up to certain critical distance (say, 2.8-3.0 A^0). Thus there is ample scope to doubt that phenomena could be propelled by the force of self gravity. Because force of self gravity increases with increase in mass. So this is a gap area of investigation.

Again 'denaturation' of protein results in disruption in cell activity and possibly can proceed up to cell death. Why cell activity is to be disrupted, or cell death should occur with simple change in physical form, if chemistry is dominating? It is interesting to note that under 'renaturation'[6], proteins can regain their native state when the denaturing influence is removed. So unless a universal binding force of self gravity is not brought into picture, native state of globular protein, 'denaturation' and again reversing to its original form of 'renaturation' will continue to remain elusive. Unless a common omnipotent force is not brought into the scene, the whole phenomena of native conformation ('naturation'), 'denaturation' and 'renaturation' of protein in a regimented manner

cannot be explained. Only common omnipotent natural force of getting united and reunited after withdrawal of external force is the self gravity under given circumstance. So it is difficult to ignore invisible force of self gravity and hence it is gap area of investigation in life science.

Spheroids fibrous protein and self gravity

Though made up of almost same amino acids, the shape of fibrous proteins, often called scleroproteins, look like a long filament or rod, and usually serve as inert structural or storage protein. Keratin, collagen, elastin, and fibroin are all scleroproteins. Their role is limited to protection and support, forming connective tissue, tendons, bone matrices, and muscle fiber. Most of its polypeptide chain is parallel to a single axis and are often mechanically strong & highly cross-linked. Under classic artefactual cell culture conditions in a flat, rigid petri dish or as per geometry of the contact surfaces, it remains in elongated position. It could possibly withstand the force of self gravity and remain in extended structure. But the behavior of cartilage cells, for instance, could be affected significantly when they are organized in 3-D using a micropatterning technique and on carefully positioning the cells within about 10 microns of each other i.e. nearly the diameter of a cell and about one-fifth the diameter of a human hair. Though process was observed to be slow or the size and shape of the cell clumps varied significantly, it is interesting that the cells clump together into "cell spheroids"[7]. From the above it is clear that spheroid shape could be formed due to self gravity if there is free floating or free fall condition and self gravity is allowed to operate in unperturbed manner. As self gravity and sphericity are two sides of the same coin, we can assume scleroproteins remain an exception only due to circumstances of local origin. So it could remain in extended structure, unlike compact form i.e it could possibly withstand the force of self gravity. Manuel Théry[8] while reviewing micropatterning as a tool to decipher cell morphogenesis and functions, pointed out that cell microenvironment, especially positioning of adjacent cells, location and orientation of extracellular matrix (ECM) fibres imposes specific 'boundary conditions' that influence cell architecture and mechanics. The size and stiffness of the microenvironment limits cell volume and cell spreading. However for geometrical control, only few studies have combined the soft, deformable substrates as medium. Hence environment that allows the self gravity to work unperturbed needs to be understood in better manner.

Why cell density increases before growth or heavier cells grow faster?

Cell growth comprises changes in both mass and volume. Understanding relationship among the cell's three basic physical parameters viz. mass, volume, and the ratio of the two, density, is important to the study the effect of invisible force of self gravity. Using buoyant mass, growth of single cells has recently been measured. With the suspended microchannel resonator (SMR), particles are weighed in real-time as they flow through a hollow cantilever. The microchannel resonant frequency is determined by the difference in mass of the particle with respect to that of the displaced fluid. Thus, the particle's density is determined by measuring its mass in two fluids of different densities. Michel Godin et al[9] found that for individual cells of *Bacillus subtilis*, *Escherichia coli*, *Saccharomyces cerevisiae* and mouse lymphoblasts, heavier cells grew faster than lighter cells. Andrea K. Bryan et al[10] found that cell density increases prior to bud formation of the yeast *Saccharomyces cerevisiae*. To investigate the origin of this density increase, they monitor relative density changes of growing yeast cells. They focus on basic cell cycle questions in yeast, but they remain oblivious on the invisible force of self gravity. They found that the density increase requires energy, function of the protein synthesis. But they have not defined the required source of energy in appropriate dimensions. Let us analyze such energy in the perspective of self gravity.

In the earlier article 'Invisible force of self gravity- A gap area of investigation in life science' we have demonstrated that hydrostatic balance or hydrostatic equilibrium is the law of biological growth, as with stellar bodies, though materials are different. We have already assumed that bio-matters/ cells are held together by gravity that tries to compress everything to the center. But to understand how things works cell by cell i.e. layer by layer, it is required to formulate an equation of state. Density, pressure, and temperature of bio-matters comprising solids, liquids and gas are related though it is very complex and little uncertain. Normally pressure is the amount of force/area. But in equation of state, pressure may be considered as a constant (K) multiplied by the mass density and the temperature divided by the molecular weight of the bio-materials. The molecular weight is the weighted mean of the different atomic types, taking into account the relative proportions of the different types of atoms

present in the bio-matters. Mass density is the amount of mass/volume. Temperature is a measure of the random motion energy (the average kinetic energy) of the solid and liquid particles so also gas to reckon. The higher the temperature, the more random is the kinetic energy of the materials. Warmer materials expand to create pressure on its surroundings.

Materials are compressed by gravity to smaller volumes and higher densities. Deeper layers have more gravity compression from the overlying layers of cells, as density as well as temperature is increased at the core. So they have greater outward pressure to compensate. Thus under equation of state, pressure gradient force or difference in pressure exceeding hydrostatic balance or hydrostatic equilibrium across a surface in each layer of cells causes a difference in force, which can result in acceleration according to Newton's second law, if there is no additional force to balance it. The resulting force is always directed from the region of higher-pressure to the region of lower-pressure. Hence it is natural to find that increase in cell density or increase in heaviness would foster growth of the living cells. Because difference in pressure across a surface causes a difference in force directed from the region of higher-pressure to the region of lower-pressure under different density gradient. This possibly results into heavier cells to grow faster than lighter cells or cell density to increase prior to bud formation or to exhibit similar other phenomena (Illustration 2). The matter will be clearer on directing appropriate research on system biology encompassing self gravity in threadbare manner.

Why cell adjust mass-to-volume ratio?

It is worthy to note that cells adjust their mass-to-volume ratio during important processes such as cell cycle progression[11], apoptosis [12, 13], differentiation [14], disease state[15, 16], and malignant transformation[17]. In biology and medical diagnostics, correlations of mass and density with disease and other physiological states have been established, e.g. in the various stages of malaria[18]. But these cellular-level parameters remain poorly investigated, especially as a system in relation to their self-gravitating environment under the equation of state as per hydrostatic equilibrium. Cell size is fundamental to cell cycle, cell type and cell state. Variation in cell density is related to changes to rates of mass and volume accumulation. During exponential growth, cells require coordination between growth and division to maintain the population's size distribution, but it remains unclear how cells monitor and regulate cell cycle entry in response to cell size. For instance, concentration of critical regulatory proteins is considered as the key to cell cycle control. But such concentration should not be defined by expression levels only, as concentration is always dependent on volume. The volume of the cell needs to be coupled to mass and energy requirements. So as per equation of state, pressure is a constant (K) multiplied by mass density and temperature divided by molecular weight of the materials. Mass density is the amount of mass/volume. Any variation in mass density i.e. mass or volume will alter the equation of state. This could result into different cell type, cell density and cell cycle. These are not yet investigated in light of self gravitating environment. Hence this is a gap area of investigation.

William H. Grovera et al[19] had developed technique to measure single-cell mass, volume, and density. They had attempted to demonstrate this technique with four examples: identifying *Plasmodium falciparum* malaria infected erythrocytes in a culture, distinguishing transfused blood cells from a patient's own blood, identifying irreversibly sickled cells in a sickle cell patient, and identifying leukemia cells in the early stages of responding to a drug treatment. The ability to measure single-cell mass, volume and density would provide valuable insights into cell state for a wide range of biological processes, provided such study is also oriented towards understanding the equation of state encompassing invisible force of self gravity.

Some exceptions to general rule

Molar mass and density based thumb rule for action of the self gravity in organizing macromolecules may be violated by various local forces operating at particular period of formation, especially for proteins, lipids and others. For instance, hydrogen bonding, ionic interactions, Van Der Waals forces, and hydrophobic packing often might disturb the general pattern of attraction in protein macromolecules. Amyloids[20] for instance, are insoluble fibrous protein aggregates sharing specific structural traits. They arise from at least 18 inappropriately folded versions of proteins and polypeptides present naturally in the body. These misfolded structures alter their proper configuration such that they erroneously interact with one another or other cell components forming insoluble fibrils. They have been associated with the pathology of more than

20 serious human diseases in that, abnormal accumulation of amyloid fibrils in organs may lead to amyloidosis, and may play a role in various neurodegenerative disorders. The site specific local environments of these proteins are required to be defined from the point of surrounding medium or metabolically inert infrastructure, apart from mass, volume and density of the cell or protein. The idea can be clear from the following facts.

Behavior of living mass under different types of medium

It is to be remembered that fluids play an important role towards buoyant like physical force that separate self gravitating biomass in living body from the surrounding inertial gravitational forces that might perturbed the action of self gravity. In addition to cytoplasmic and other fluid matrix in living cells, human body contains various fluids like blood plasma, lymphatic fluid, interstitial fluid, viscous fluid of mucus, saliva, gastric juice, cerebrospinal fluid, sweat, tears, the aqueous and vitreous humors of the eye, semen, vaginal secretions, amniotic fluids etc. The composition, density and depth of each fluid differ affecting operational buoyant force. For instance, brain exists in neutral buoyancy. The actual mass of the human brain is about 1400 grams; however, the net weight of the brain suspended in the CSF under neutral buoyancy is equivalent to a mass of 25 grams. Similarly blood cells are suspended in a fluid called blood plasma, which is mainly composed of water and a mixture of other dissolved substances, or solutes apart from hormones, vitamins, amino acids, and antibodies. Blood plasma has a density of approximately 1.025 kg/l. In comparison to plasma, glucose in cerebrospinal fluid (CSF) is diminished by about 80%. It is a gap area of investigation that how far disturbance in operational buoyant force can perturb various activities of life processes.

Cells traditionally have been studied in two dimensions (2-D) in a petri dish, but certain cells behave differently in three dimensions than in two. Size and shape of the cell depend on the external biophysical forces. MIT bioengineers[7] have provided pictures that show normal and diseased cartilage cells which are organized differently in normal and diseased cartilage and 3-D cell clusters of same normal and diseased cartilage precisely re-created in a tissue like gel compared to cells in a conventional 2-D petri dish (Illustration 3). External biophysical force is to be examined critically for specifying behavior of the living mass on it, especially gravitating environment.

Importance of medium vis-a-vis unperturbed self gravity

We have assumed[1] that self-gravitating biomass/ embryo is in the accelerated reference frame, manifesting its physiological and genetic functionality. ‘Metabolically Inert Infrastructure (MII)’ is placed in the co-moving non-accelerated reference frame that are relatively stationary or at constant velocity, or non-aligned or acting in opposite direction of the energized accelerated self gravitating biomass or of the steady state supporting inertial reference frame at the specific point of time. Let us see how it applies in case of mediums required as microbial or biotechnological analysis protocols like agarose, polyacrylamide, silica colloidal crystal (SCC), raffinose etc. For instance, three “blot” techniques are utilized for detecting presence and relative quantities of specific macromolecules (DNA, RNA, protein) in cells viz. DNA (Southern) with agarose/ acrylamide, RNA (Northern) with agarose, and protein (Western) with polyacrylamide. Why to be positioned over agarose or other gel? The agarose gel is a cross-linked matrix that is somewhat like a three-dimensional mesh or screen[21]. When boiled agarose cools, it forms a loose molecular net resembling a sponge with required mechanical rigidity in soft porous texture. The pores in the gel matrix are filled by the liquid phase. Buoyant like force of the liquid is thus augmented by mechanical rigidity of the surrounding structure. Thus apart from other known advantages, final agarose gel gets the ability to withstand compressibility and allows the positioned biomass to feel less stressed under concentrated gravitational load (illustrations 4). In Southern blotting, for instance, after separation of fragments according to length, a sheet of either nitrocellulose paper or nylon paper is laid over the gel, and the separated DNA fragments are transferred to the sheet by blotting. The gel is supported on a layer of sponge in a bath of alkali solution, and the buffer is sucked through the gel and the nitrocellulose paper by paper towels stacked on top of the nitrocellulose (Illustration 5). As the buffer is sucked through, it denatures the DNA and transfers the single-stranded fragments from the gel to the surface of the nitrocellulose sheet, where they adhere firmly. Stress applied from own weight and from external load is known as effective and net stress respectively. The

bulk modulus of a substance, on the other hand, measures the substance's resistance to uniform compression or stress. For effective rafting, the biomass is required to be isolated or free from stress on flotation or through other mechanisms. Stress applied from own weight differs among RNA, DNA and protein fragments due to obvious reason of difference in molar mass and density. Also agarose gels have larger 'pores' than polyacrylamide gels meaning that it packs less densely then an equivalent amount of polyacrylamide. Therefore, considering variation in packing density, agarose is generally used for the electrophoresis of large molecules such as DNA and RNA and polyacrylamide is used for small molecules such as proteins. Uneven local mass distribution causes local variation in density as well as gravitational attraction. Accordingly choice is being made among various available materials.

Evans et al[22] pointed out that while cell attachment was unaffected by the stiffness of the growth substrate, cell spreading and cell growth were all increased as a function of substrate stiffness and the mechanical environment can play a role in both early and terminal embryonic stem cells (ESC) differentiation. Ji L et all[23] found that cells cultured on the substrates formed of silica colloidal crystal (SCC) retained transcription of stem cell and endoderm markers more similar to undifferentiated ESCs, suggesting the substrates are restricting differentiation, particularly towards the endoderm lineage, compared to cells cultured on flat glass. Additionally, five days after seeding, they observed strikingly different colony morphology, with cells on the SCC substrates growing in spherical colonies approximately ten cells thick, while cells on glass were growing in flat monolayers. Colonies on the SCC substrates developed a central pit, which was never observed in cells cultured on glass, and expressed proteins related to epithelialisation. Together, these data demonstrate the potential of using topographical cues to control stem cell behaviour in vitro. For smallest prion, say ([Het-s] prion (molecular weight of 35-36kDa) of the filamentous fungus *Podospora anserina* transformants are grown in liquid raffinose synthetic medium (SR) plus galactose. As stated above, molar mass of raffinose, a trisaccharide composed of galactose, fructose, and glucose is 504.42 g/mol with density 1.723 g/cm^3. Therefore it seems that medium plays a vital role towards unperturbed action of self gravity. For smallest prion, a protein, liquid reffinose, a carbohydrate is sufficient to increase density of the liquid medium and thereby provide a separation due to pressure of up thrust from inertial gravity, where as for DNA, RNA, larger proteins, whose molecular weight and density is comparatively higher, mechanical prop up support from agarose gels etc. is additionally required. However all these are gap areas of investigation.

Concluding remarks:

The purpose of the study was to identify various gap areas of life science towards invisible force of self gravity. Biological science was so far silent on the existence of self gravity. But all those evidences[1] in addition to those narrated in the present article, starting from macromolecules to organism level, it is apparent that self gravity plays a dominant role in the functioning and manifestation of various walks in the journey of an organism. Hence appropriate study needs to be directed towards such unexplored field encompassing 'Self gravitation bio' or 'Biomechanics of intrinsic gravity'. Present sporadic studies may not be sufficient for full length explanations of many of the biological phenomena, but complexity and multitude of mysteries in diverse biological sciences can not be solved in a single stroke. Here one must accept that 'direction'is more important than 'distance'. Also it is to be remembered that various unusual phenomena encountered in space biology under microgravity environment can also posibly be resolved satisfactorily with the idea of self gravity, that would be clear from the narration to be made in the forthcoming article on interaction of self gravity with extrinsic inertial gravity.

References

1. Bhattacharjee IR. Invisible Force of Self Gravity: A Gap Area of Investigation in Life Science . WebmedCentral BIOPHYSICS 2012;3(9):WMC003672 http://www.webmedcentral.com/article_view/3672

2. Hannes Fischer,Igor Polikarpov,and Aldo F. Craievich. Average protein density is a molecular-weight-dependent function. Protein Sci. 2004 October; 13(10): 2825–2828. doi: 10.1110/ps.04688204 PMCID: PMC2286542

3. http://es.wikidoc.org/index.php/Communal_aggregation

4.http://www.britannica.com/EBchecked/topic/479680/protein/72530/The-isolation-and-determination-of-proteins

5.

http://en.wikipedia.org/wiki/Denaturation_(biochemistry)
6. http://www.blurtit.com/q932058.html
7. http://web.mit.edu/newsoffice/2006/3d.html
8. Manuel Théry. Micropatterning as a tool to decipher cell morphogenesis and functions. December 15, 2010 J Cell Sci 123, 4201-4213. doi: 10.1242/jcs.075150
9. Michel Godin , Francisco Feijó Delgado, Sungmin Son ,William H Grover , Andrea K Bryan , Amit Tzur , Paul Jorgensen , Kris Payer , Alan D Grossman , Marc W Kirschner & Scott R Manalis. Using buoyant mass to measure the growth of single cells Nature Methods 7, 387–390 (1 May 2010) | doi:10.1038/nmeth.1452
10. Andrea K. Bryan, Alexi Goranov, Angelika Amon, and Scott R. Manalis. Measurement of mass, density, and volume during the cell cycle of yeast. www.pnas.org/cgi/doi/10.1073/pnas.0901851107 PNAS ? January 19, 2010 ? vol. 107 ? no. 3 ? 999–1004
http://www.pnas.org/content/107/3/999.full.pdf
11. Wolff DA, Pertoft H (1972) Separation of HeLa cells by colloidal silica density gradient centrifugation. I. Separation and partial synchrony of mitotic cells. J Cell Biol 55:579–585.
12. Martin SJ, Bradley JG, Cotter TG (1990) HL-60 cells induced to differentiate towards neutrophils subsequently die via apoptosis. Clin Exp Immunol 79:448–453
13. Wyllie AH, Morris RG (1982) Hormone-induced cell death. Purification and properties of thymocytes undergoing apoptosis after glucocorticoid treatment. Am J Pathol 109:78–87.
14. Maric D, Maric I, Barker JL (1998) Buoyant density gradient fractionation and flow cytometric analysis of embryonic rat cortical neurons and progenitor cells. Methods 16:247–259.
15. Mrema JE, Campbell GH, Miranda R, Jaramillo AL, Rieckmann KH (1979) Concentration and separation of erythrocytes infected with Plasmodium falciparum by gradient centrifugation. Bull World Health Organ 57:133–138.
16. Rodgers GP, Schechter AN, Noguchi CT (1985) Cell heterogeneity in sickle cell disease: Quantitation of the erythrocyte density profile. J Lab Clin Med 106:30–37.
17. Bosslet K, Ruffmann R, Altevogt P, Schirrmacher V (1981) A rapid method for the isolation of metastasizing tumour cells from internal organs with the help of isopycnic density-gradient centrifugation in Percoll. Br J Cancer 44:356–362.
18. Mills JP, et al. (2007) Effect of plasmodial RESA protein on deformability of human red blood cells harboring Plasmodium falciparum . Proc Natl Acad Sci USA 104:9213–9217.
19. William H. Grovera,Andrea K. Bryana, Monica Diez-Silva, Subra Suresh, John M. Higgins, and Scott R. Manalis. Measuring single-cell density. May 23, 2011 www.pnas.org/cgi/doi/10.1073/pnas.1104651108 PNAS Early Edition ? 1 of 5.
20. http://en.wikipedia.org/wiki/Amyloid
21. Laas, T. Gel structure of agarose. Doctoral thesis. Acta Universitatis Upasaliensis 1975. http://students.washington.edu/uwfarm/2011/02/10/food-science-2-0-fragments-of-heredity/
22. Evans ND, Minelli C, Gentleman E, LaPointe V, Patankar SN, Kallivretaki M, Chen X, Roberts CJ, Stevens MM. Substrate stiffness affects early differentiation events in embryonic stem cells. Eur Cell Mater. 2009 Sep 21;18:1-13; discussion 13-4. http://www.ncbi.nlm.nih.gov/pubmed/19768669
23. Ji L, LaPointe VL, Evans ND, Stevens MM. Changes in embryonic stem cell colony morphology and early differentiation markers driven by colloidal crystal topographical cues. Eur Cell Mater. 2012 Feb 23;23:135-46.
http://www.ncbi.nlm.nih.gov/pubmed/22370796
24. Vidhu Mathur, Vibha Taneja, Yidi Sun, Susan W. Liebman. Analyzing the birth and propagation of two distinct prions, [PSI+] and [Het-s]y, in yeast. 2010. Cell Biology of Disease. http://www.molbiolcell.org/content/21/9/1449.full

Illustrations

Illustration 1

Illustration 1. Self organization of macromolecules in living cell: Force of self gravity attracts nucleic acids to cell's near central position due to highest molar mass and density. Proteins have intermediate molar mass- thus remain in intermediate position. Fats and lipids are less dense than others remain in periphery in cell membrane. Carbohydrates have higher solubility and contribute fluids, buoyant force on which assists to provide free floating (free fall) condition.

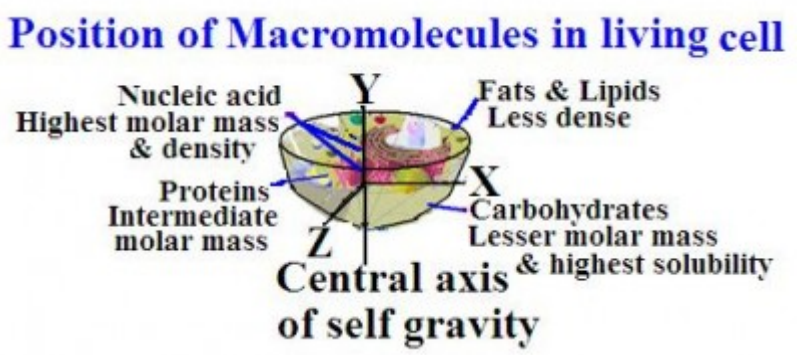


Illustration 2

Illustration 2. Due to hydrostatic balance or hydrostatic equilibrium (gravity versus internal pressure) under equation of state, difference in pressure across a surface under different density gradient causes a difference in force directed from the region of higher-pressure to the region of lower-pressure. This possibly results into heavier cells to grow faster than lighter cells or cell density to increase prior to bud formation or to exhibit similar other phenomena.

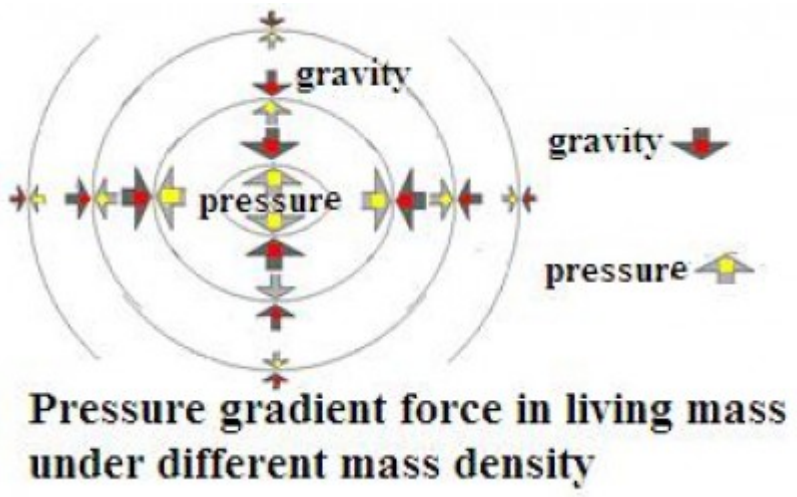


Illustration 3

Illustration 3. Effect of extrinsic inertial gravitational force and unperturbed self gravity: MIT bioengineers7 have provided pictures that show normal (above left) and diseased (above right) cartilage cells which are organized differently in normal and diseased cartilage and 3-D cell clusters of same normal (middle left) and diseased (middle right) cartilage precisely re-created in a tissue like gel compared to cells (bottom) in a conventional 2-D petri dish.

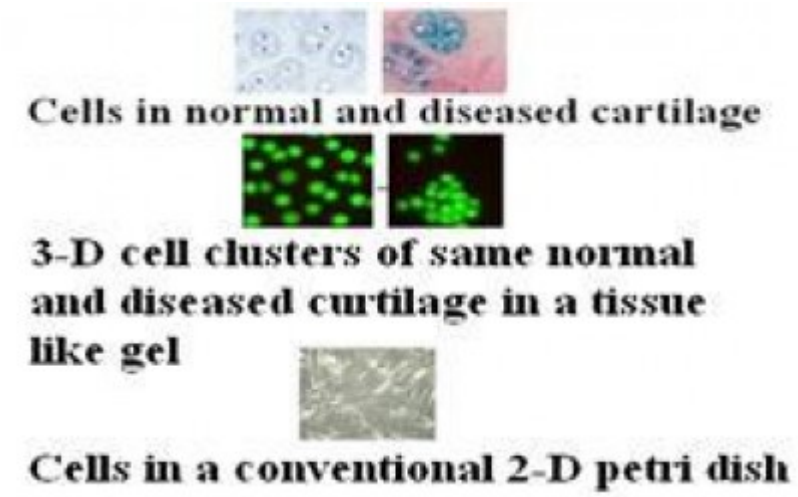


Illustration 4

Illustration 4. Final cross linked gel structure matrix of agarose allows effective stress distribution of concentrated gravitational load

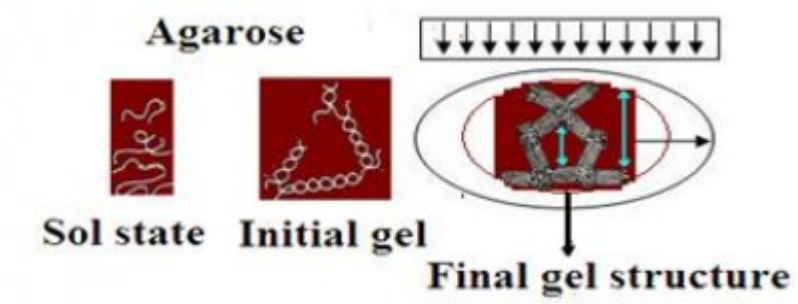
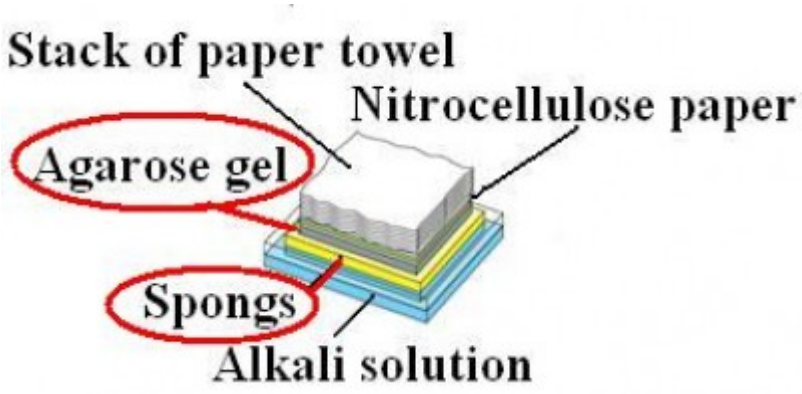


Illustration 5

Illustration 5. Why bio-matters are to be positioned over sponge and agarose gel? Sponge as well as agarose gel provides mechanical rigidity in order to withstand compressibility or bulk modulus (substance's resistance) of the stress applied from own weight (effective stress) and from external load (net stress). For effective rafting, the biomass is required to be isolated or free from stress on flotation or through other mechanisms.



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