Indian J.Sci.Res. 12 (2): 128-133, 2017

ISSN: 0976-2876 (Print)
ISSN: 2250-0138(Online)

SYNTHETIC BIOLOGY: A NOVEL BIOINSPIRED APPROACH TO EMERGENCE OF LIFE: PHOTOCHEMICAL FORMATION OF BIOMIMETIC PHOTOAUTOTROPHIC ASSEMBLIES "JEEEWANU" IN A LABORATROY SIMULATED POSSIBLE PREBIOTIC ATMOSPHERE

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ABSTRACT

Sunlight exposed sterilized aqueous mixture of ammonium molybdate, di ammonium hydrogen phosphate, biological minerals and formaldehyde shows photochemical formation of biomimetic, self-sustaining protocell-like supramolecular assemblies, "Jeewanu" [1]. The microscopic examination of Jeewanu have revealed that they are spherical in shape and are capable of showing multiplication by budding, grow from within and metabolic activities in them. In prebiotic atmosphere possibly photosynergistic collaboration of non-linear processes at mesoscopic level established autocatalytic pathways on mineral surfaces by self-organisation,led to emergence of supramolecular photoautorophic assemblies similar to Jeewanu which might have given rise to earliest energy transducing common universal ancestor on the earth or elsewhere.

KEYWORDS: Jeewanu, selforganisation, autocatalytic, protocell-like assemblies, biomimetic, supramolecular, photoautrophic, bioinspired

One of the most fundamental problems of origin of life is that in primitive atmoshphere how energy transferring systems would have converted sunlight into chemical form. Lipman [1] postulated that in all cells a tendency exists to convert major part of oxidation-reduction energy into phosphate bond energy. Living organisms photochemically trap energy by the following two mechanisms.

i.Photochemical reduction of CO2

ii. Photophosphorylation

The concept of photosynthetic phosphorylation suggested by Arnon et.al [2] opened up new vistas in the mechanism of conversion of solar energy into energy rich biological compounds and in electron transport phosohorylation.

ENERGETIC COUPLING

Chemically Intelligible Reactions

Several cases of phosphorylation viz. non-phosphorylation [3, 4, 5, 6]; inorganic photophosphorylation [7, 8]; organic photocatalysis [9, 10] have been studied by various workers. The direct photophosphorylation of ADP with iP ^{32} to ATP in aqueous suspension of semiconductors has been reported [11, 12]. An extremely sensitive methods for PPi synthesis was suggested by Nyren & Ludin (13). Bacteriorhodopsin is a light driven photon pump [14]. The energy absorbed by them is utilized to drive ATP synthtase which converts ADP and inorganic phosphate into ATP [15]. The metabolic production of ATP can be viewed as a general mechanism for the coupling of energy yielding end energy requiring process. Coupling of metabolic activities and functional activities. The generation of ATP by chromatophores of photosynthetic synthetic bacteria has been investigated [16]. In chromatophores of R.rubrum the driven transport is catalysed by alternating phosphorylating enzymes, the proton translocating PPase, which both are membrane bound [17]. It was observed that final product of energy transfer reactions PPi and ATP respectively are formed at a catalytic site or very close to the outer membrane of the chromatophores [18].The concept of phosphorylation introduced entirely new possibilities based on light energy.

PHOTOSYNTHETIC APPARATUS

A Supramolecular Array

In photobiological systems the phtotosynthetic apparatus consist of macromolecules (proteins embedded in a matrix of a bilayer phospholipids membrane [19]. The early events in photosynthetic energy conversion include phto-induced electron transfer mediated by donor and

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acceptor moieties. The small organic species are bound to proteins, which in turn are embedded in a lipid bilayer membrane. Thus the photosynthetic apparatus of a plant or other organisms actually comprises a large supramolecular array. In these covalent linkages model the structural role of proteins [20]. The emergence of chemistry beyond molecules [21] has initiated a shift over last 25 years where concept like self-assembly are transferred from biological processes into chemical nanosystems through the medium of syntheses. Construction of natures molecules in the laboratory from atoms or single molecules a process known as “Total Synthesis” [22]. Molecular self-assembly, a concept central to nature’s form and functions [23]. Assembly of molecular components to obtain photochemical devices has been studied [24, 25]. Supramolecular chemistry demonstrates cooperativity at structural and functional level.

At present best –characterised biological motor is ATP synthtase. The synthesis of ATP is based on a proton pump across a membrane. Perhaps the most spectacular molecular machine constructed in recent years is a biomimetic power ATP synthtase to produce ATP [26]. The working mechanisms of key biological machines that involved ATP syntheses have been studied in detail [27, 28]. Balzani studied Photo induced electron transfer to chemical potential associated with ATP-ADP conversion. It constituted a synthetic biological motor or a biomimetic system [29].

The photochemical formation of protocell-like microstructures “Jeewanu” was observed in a sunlight exposed sterilized aqueous mixture of ammonium molybdate, diammonium hydrogen phosphate, biological minerals and formaldehyde [30, 31, 32]. These microstructures have a definite boundary wall and intricate internal structure. They multiply by budding, grow from within by actual synthesis of material and are also capable of showing various metabolic activities [31]. Jeewanu have been analysed to contain a number of biochemicals-like materials in them viz. amino acids which are present in free as well as in peptide combination, nucleic acid bases as purines as well as pyrimidines, sugars as ribose as well as de-oxyribose and phospholipids-like material in them. The presence of urease, esterase, peroxidase and phosphatase –like activities have been detected in the mixture [32, 33, 34]. The esterase and phosphatase-like activities in Jeewanu mixture were also reporte [32, 35]. The presence of acid phosphatase-like activity in Jeewanu has been histochemically demonstrated [36]. Jeewanu have been also found to contain ferredoxin-like material in them [37, 38]. The photochemical reduction of acetylene by Jeewanu, indicated the presence of nitrogenase-like activity in the mixture [39]. The cytochemical and histochemical investigations of Jeewanu showed that they can be fixed with biological fixatives and can be stained with acidic and basic dyes [40, 41]. It is quite possible that earliest energy transducing systems were possibly a photoautotroph. Therefore an attempt was made to investigate the photochemical formation of protocell-like microstructures Jeewanu [32] under highly precise laboratory conditions.The morphological characteristics of Jeewanu were investigated using optical, electron microscope (SCM & TEM) and Atomic Force Microscope to undertand self-organisation of photoproducts at mesoscopic level and emergence of protocell-like supramolecular assemblies “Jeewanu” in the mixture. Further an attempt was made to study whether the earliest energy transducing systems could utilize energy rich compounds like ATP. A comparative study of high and low mineral mixture of Jeewanu was carried out to find out probable primitive pathways of energy production in the laboratory simulated possible primitive atmosphere.

EXPERIMENTAL

The following two types of Jeewanu mixtures were prepared [32].

- Low Mineral Jeewanu Mixture
- High Mineral Jeewanu Mixture

Method of preparation of Low Mineral Jeewanu Mixture

The following solutions were prepared:-

- 4% Ammonium Molybdate (w/v)
- 3% Di-ammonium hydorogen phosphate (w/v)
- Mineral Solution

It was prepared by dissolving 20 mg each of potassium di-hydrogen phosphate, calcium acetate, sodium chloride, potassium sulphate, magnesium sulphate and 50 mg of ferrous sulphate in 100 ml of distilled water. The salts were added one by one, a new salt was added when one was dissolved completely by shaking.

Ammonium molybdate 50 ml (1 vol.), di-ammonium hydrogen phosphate 100 ml. (2 vol.), mineral solution 50 ml (1 vol.) were mixed in a conical flask. The flask was cotton plugged and sterilized in an autoclave at 15 lb pressure for 30 minutes. After cooling 10 ml (1 vol.) 36%formaldehyde was asceptically added in the mixture. A part of the mixture was taken in a conical flask covered with black cloth was kept as control mixture. The mixtures were exposed to sunlight for 24 hours giving 6 hours exposure each day.

Morphometric characterization of Jeewanu by optical Microscope

A drop of suspension of suspension of mixture was examined under optical microscope at 1500 X. The images obtained were digitally recorded and analysed by Image Analysis Software Pro C provided by Olympus. The various morphometric measurements were taken to characterize the photoproduct synthesized in the mixture. (Figure.1, 2)

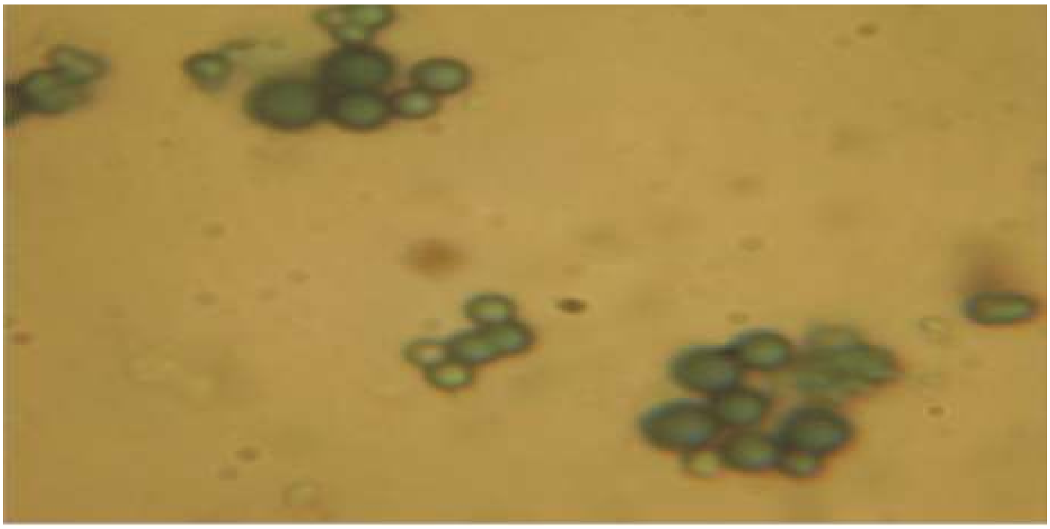

Figure 1: Low Mineral Jeewanu (1500X) showing multiplication by budding and growth from with in

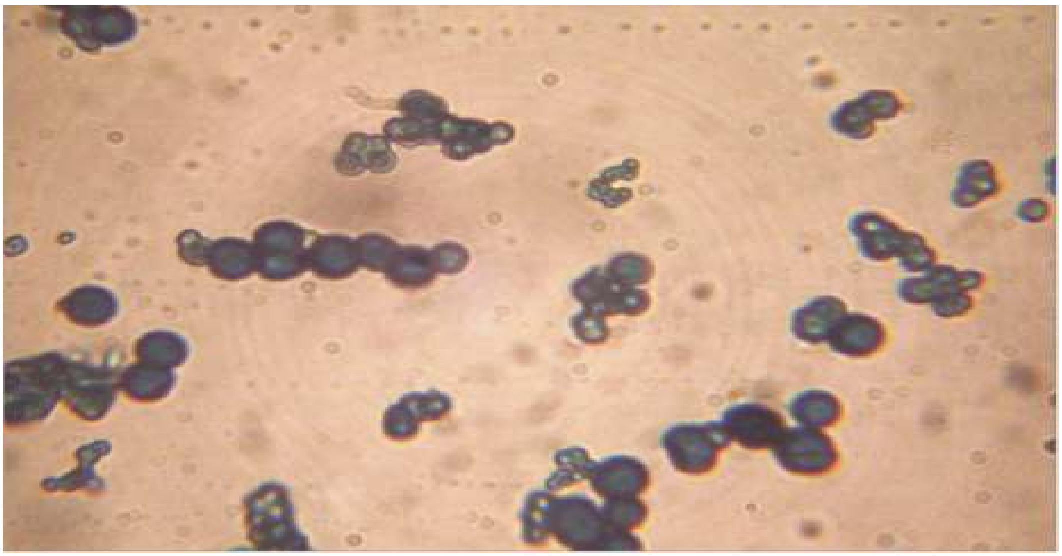

Figure 2: High Mineral Jeewanu (1500X) showing multiplication by budding and growth from with in

RESULTS AND DISCUSSION

Irradiated sterilized aqueous mixture of ammonium molybadate, diammonium hydrogen phosphate biological minerals and formaldehyde (32) shows photochemical formation of protocell-like supramolecular assemblies “Jeewanu” having a definite boundary wall and an intricate internal structure. They are capable of showing multiplication by budding, grow from within by actual synthesis of material and show various metabolic activities in them. The optical and electron microscopic studies have clearly revealed that newer smaller units comes from the parental unit by budding. The photochemical formation of Jeewanu is a self-sustaining process and are formed by autocatalytic photochemical transformations mediated by inorganic metal ions and transitional elements present in the mixture. The scanning probe microscopy (SPM) of Jeewanu revealed their structural characteristics at high resolution showing the presence of microstructure in different stages of their formation. The optical microscopic and transmission microscopic studies have shown that Jeewanu are spherical in shape, have a definite boundary wall and an intricate internal structure.

CONCLUSION

In primitive atmosphere possibly photosynergistic collaboration of non-linear processes at mesoscopic level led to selforganisation and emergence of supramolecular self-sustaining assemblies similar to “Jeewanu”. The systems chemistry concerning formation of dynamic covalent bonds, quantum mechanical resonance stability force and electromagnetic interactions must have led to spatio-temporal coherence showing a cooperative informational hierarchy between structure and function. The formation of non-covalent bond and spontaneousself-organisation of membrane (46) is of much interest. Cairns Smith (47) postulated that the first photosynthetic systems would have been made of clays such mineral membranes could hold transitional metal ions to catch light and conduct charges as well as inert barrier to separate photoproducts.

It can be said that energy rich compounds like ATP were synthesized in the prebiotic atmosphere by photophosphorylation reactions and were degraded to ADP and Pi with the help of ATP-ase-like enzymes. Energy thus released was utilized various bioenergetic processes. Presence of ATP-ase-like activity in Jeewanu mixture suggests that earliest energy transferring system would have been of jeewanu grade of organization.

The free energy is needed to overcome thermodynamic limitations. In prebiotic atmosphere possibly photo-isomerisation and conformational changes initiated novel coherent emergent phenomenon at mesoscopic level, subsequently evolution of common universal ancestor or open chain microstructures possibly similar to Jeewanu showing certain degree of intelligence.

ACKNOWLEDGEMENT

Author gratefully acknowledges University Grants commission, CRO, Bhopal (M.P.) India for the grant no. MRP/MS-69/201001/XII/14-15/CRO/ 02-03-2015 Dr D.A. Lerner, Professor Emeritus, Ecole Nationale De Chimie, Montpellier, France for the valuable cooperation and discussions.

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