

Cytoplasmic metabolites study of Vegetative and Reproductive structures of *Solieria robusta*

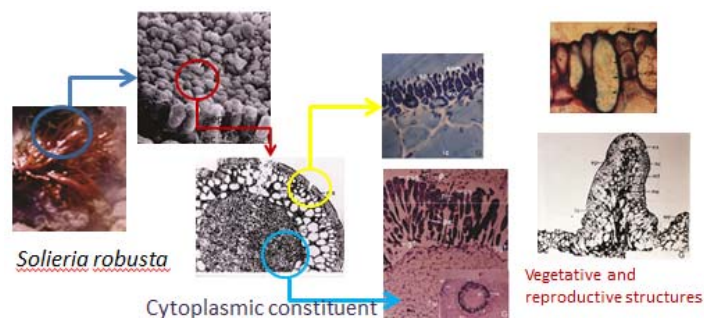
Indu Nashier Gahlawat*

Department of Biology, Aditi Mahavidyalaya, University of Delhi, Bawana, Delhi - 110039 India.

Received on: 25-Mar-2020, Accepted and Published on: 16-May-2020

ABSTRACT

Solieria robusta exhibits triphasic isomorphic alternation of generation. The plants of the male, female and tetrasporophytes are terete with erect lateral branches that show attenuation at their bases. The thallus is differentiated into cortex and medulla. Spermatangial mother cell and spermatangial cytoplasm stain moderately for sulphated and carboxylated polysaccharides whereas mature tetrasporangia, tetraspores, and carpospores are rich in sulphated polysaccharides. The tetrasporangial cytoplasm like carposporangial cytoplasm show fibrous vesicles, chloroplast, mitochondria and floridean starch grains. A thick electron transparent mucilaginous sheath covers the tetrasporangial wall whereas carposporangial wall is 3 to 5 layered. The carpospores and tetraspores are replete with floridean starch grains which are of various shapes and size. *Solieria robusta* (Greville) Kylin shows regeneration.



Keywords: Red Algae, Histochemistry, Metabolites, Seaweed, Regeneration

INTRODUCTION

Indian resources in seaweeds, are sufficiently valuable but they are not fully explored and utilized. Sea weeds are important sources of alginic acid (alginates), agar and carageenans which are useful in food, textile and pharmaceutical industries. With depleting land and increasing population, one has to look towards the marine ecosystem for food and survival. Almost half the Florideophyceae families belong to the Gigartinales one of the most diverse orders of the Florideophycideae. In Gigartinales the auxiliary cell is produced in the vegetative filament, Red algae cell wall contain copious amount of carboxylated and sulphated polysaccharides. These polysaccharides can be extracted from many seaweeds which grow luxuriantly in subtidal regions. To make this approach feasible, it is necessary to investigate the reproductive biology of these taxa using modern methods. Histochemistry helps in situ localization of important metabolites during progressive

developmental stage in the life cycle. Electron microscopic studies help in revealing better resolution of cell organelles, relative distribution and their probable function. The red algae *Solieria robusta* grows in rock pools in Okha coast. The earlier reports on this taxon casts many doubts and lacunae in our knowledge with respect to its origin, development and ultrastructure of vegetative and reproductive thalli. The information on reproductive thalli, reproduction biology, particularly a correlative account of developmental, histochemical and ultrastructural aspects is very less. The histochemical studies detect all changes that occur in the wall polysaccharides and cytoplasmic metabolites during progressive stages of development of both vegetative and reproductive structures. The present work is aim to study histochemical details that occurs in the wall polysaccharides and cytoplasmic metabolites during progressive stages of development of both vegetative and reproductive structures. Detailed study of Carposporogenesis and tetrasporogenesis at both light and electron microscopic level is undertaken.¹⁻⁴ This work on *solieria robusta* confirms that it is non-procarpic and show process of wound generation and add on a complete new aspects to our knowledge about *solieria robusta*.

MATERIAL AND METHODS

Red Alga *Solieria robusta* (Greville) Kylin was collected at Port Okha(Gujarat) during the low tide periods for light microscopic studies, the parts of the thalli were observed under stereomicroscope

*Corresponding Author: Dr. Indu Nashier Gahlawat
Associate Professor, Aditi Mahavidyalaya, University of Delhi,
Bawana-110039, Delhi, India.
Tel: 9811241935
Email: induingahlawat@gmail.com

Cite as: *J. Int. Sci. Technol.*, 2020, 8(2), 21-30.

©IS Publications ISSN: 2321-4635 <http://pubs.iscience.in/jist>

and also processed for light microscopic studies. Two-micron thick sections were cut with glass knives using a locally made adaptor that fits into the rotary microtome. The sections are stained with 0.05% Toluidine Blue O (TBO) at PH 4.4; Periodic - acid Schiff's reagent (PAS) and Coomassie Brilliant Blue (CBB).10 Photomicrographs were taken on ORWO B/W film using Reichert Polyvar photomicroscope.

For transmission electron microscopy (TEM), parts of thalli were fixed in 6% glutaraldehyde prepared in 0.02M phosphate buffer at pH 6.8 and post fixed in 1% osmium tetroxide in the same buffer. The tissues were dehydrated in ascending aqueous ethanol and propylene oxide series. Infiltration was done in Epon-Araldite mixture.

ELECTRON MICROSCOPY

A Ultrathin sections were cut on Reichert Ultramicrotome using glass knives. Staining was done with uranyl acetate and lead citrate. Ultrathin sections were observed under Philips EM 300 electron microscope.

B For scanning electron microscopy selected sections of the thalli were passed through a graded cold (4°C) increasing acetone medium, dehydrated in anhydrous acetone, treated for critical point drying (CPD), coated with gold and observed under Philips SEM 501-B.

RESULTS

Vegetative Morphology & Anatomy

In Okha Coast (Gujarat), there is narrow supralittoral zone which is inhabited by a few green algae and molluscs (Figure 1A). Red seaweeds occur mainly in the subtidal region whereas brown and green algae are abundant in the intertidal zone. The Okha-reef is characterized by hard, corrugated, limestone rocks that are covered with calcareous deposits (Figure 1B). *Solieria robusta* is attached to the calcareous rocks and occur in the rock-pools of the intertidal region (Figure 1B). Occasionally, during high tide periods plants are washed ashore and can also be collected as drift plants. The thalli are attached to the substratum by a fibrous or discoid holdfast (Figure 1C) or the hapteron-like structure. The height of the plant is 10 - 40 cm in length. The colour of the thalli is purple to brown red. *Solieria robusta* consists of terete or slightly compressed, erect lateral branches which are generally attenuate at their base (Figure 1C-D) and tapered at their tips. Branching pattern is irregular. In male plants, the spermatangia are scattered on the surface of young branches. The female thallus surface shows many scattered cystocarps that appear as dark coloured dots (Figure 1D). Mature cystocarps are present near the base of the branch and the younger ones toward the tip (Figure 1D). The development of the cystocarp is thus acropetal. Scanning electron micrograph of female plant surface shows a very thick cuticle which covers the epidermal cell walls (Figure 2 C). The thallus surface is raised due to presence of cystocarp with a small circular ostioles in their centre. In male plants, the mucilage covering of epidermal cells lyse after the cutting of spermatangia (Figure 2A). The thalli surface show growth of many epiphytes notably *Polysiphonla*, *Ceramium* sp. and a number of diatoms. **APEX** : The apices in gametophytes and tetrasporophytes possess a group of apical cells (Figure 3A). The apical cell initially multiplies obliquely producing a subterminal cell (Figure 3A) which cuts off a periaxial cell (Figure 3A). A single

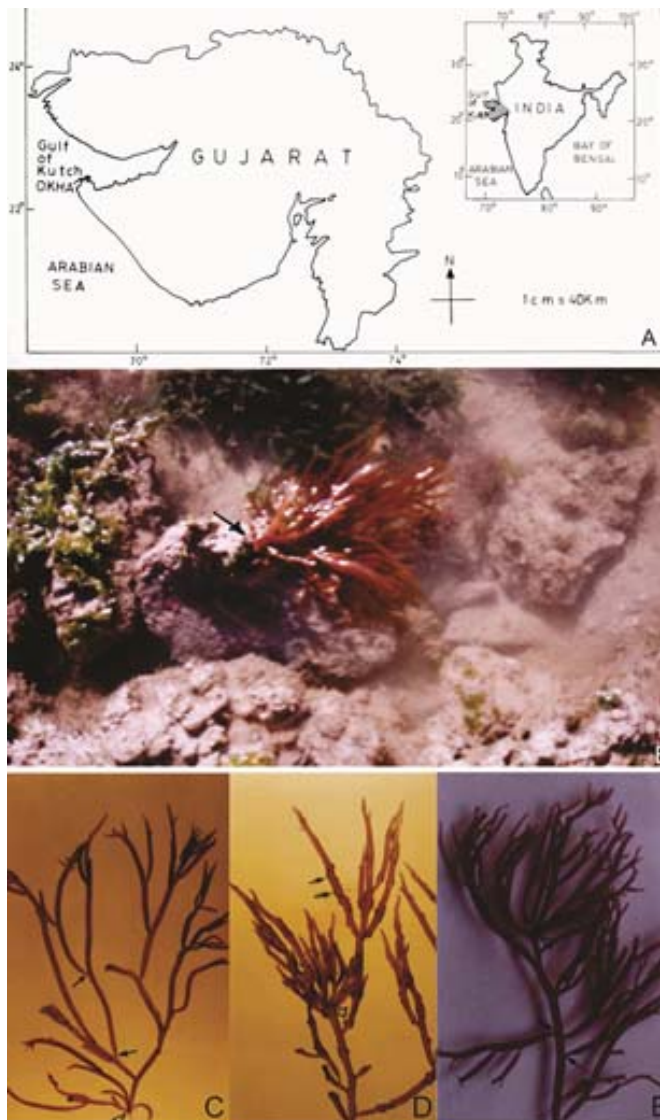


Figure 1. *Solieria robusta* morphology. (cys, cystocarp; eps, epiphytes; h, holdfast) (A) Port Okha, Gujarat, India. (B) A portion of intertidal pool to show *Solieria robusta* plant attached to calcareous rock (C) A male plant with hapteroid holdfast (Arrowhead), The branches show characteristic attenuation. (D) A female plant with swollen cystocarps (arrows). The branching is irregular and branches show attenuation at their base (double arrow). Occasionally, 4 or 5 branches arise from the damaged branch and form an umbel (arrowhead). (E) The tetrasporic plant shows attenuation of branches at the base (arrows) and formation of an umbel (arrowhead). Part of figure reproduced from ref. 1.

periaxial cell is cut off from each axial cell and succeeding periaxial cells rotate in a snaky pattern by the side of the axial filaments (Figure 3A). Periaxial cells produce lateral filaments that form the cortex. Derivatives of the apical cells become the axial filaments which later form the medulla. **AXIS** : The axis is differentiated into cortex and medulla. The cortical region comprises the epidermis, outer and inner cortices. The epidermis is unlayered (Figure 2B,C,D). The cortex is 4 to 6 cells thick which is divided into outer and inner cortex (Figure 2B,CD). The former is made up of small and the latter of big cells (Figure 2D).



Figure 2. *Solieria robusta*. Thalli surface, Gametophyte, Carposporophyte, surface view and Anatomy, SEM. (csg, carposporangium; cu, cuticle; cys, cystocarp; ep, epidermis; fu, fusion cells; fs, floridean starch grain; gf, gonimoblast filament; ic, inner cortex; me, medulla; mf, medullary filament; oc, outer cortex; p, pericarp; pc, pit connection; spm, septal plug; sp, spermatangia). (A) portion of male plant thallus surface to show that the epidermal cells are covered by mucilage covering (double arrow). Gelatinization of mucilage covering (arrows) aids in spermatia release. x 7000. (B) Sections of thalli to show epidermis and outer cortices. The epidermal cells are compactly arranged and show pit-connections with prominent septal plugs. Floridean starch grains are present in the outer cortical cells. A x 3500. (C) Longitudinal sections of thalli showing thick cuticle that veneers the epidermal cells. The outer cortical cells are gorged with floridean starch grain. X 7000. (D) Transverse section of thallus of gametophyte plant to show epidermis, outer and inner cortices. The epidermis is overarched by a thick cuticle. (arrow). The cells of cortex are replete with floridean starch grains. X 7000. (E) Longisection of a cystocarp showing 3 or 4 cells thick pericarp, fusion cells, gonimoblast filament and carposporangia. X 3500. (E) Longisection of a cystocarp showing 3 or 4 cells thick pericarp, fusion cells, gonimoblast filament and carposporangia. X 3500.

In mature thallus the cell walls of the cortical cell are thick (Figure 2D) and possess pit-connections (Figure 2B). Epidermal cells also show pit-connections with prominent septal-plugs (Figure 2B). The cells of the outer cortex are small in size as compared to inner cortex cells and are gorged with floridean starch grains (Figure 2D). The size and shape of the starch grain vary but most of them appear spherical. The tetrasporophyte thallus reveals identical anatomy. In main axis, the cortical cells show wall thickening that aids in mechanical function (Figure 2B,C,D).

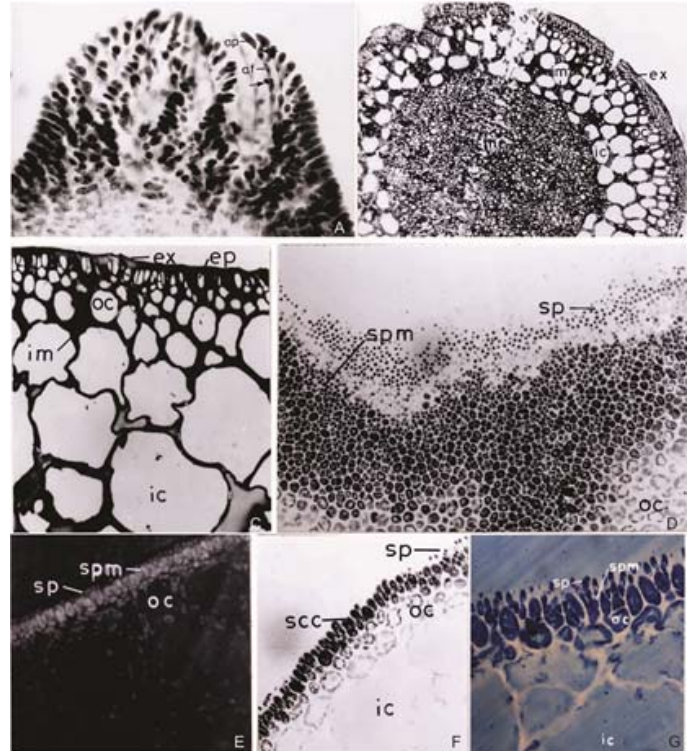


Figure 3. *Solieria robusta*. Gametophyte thallus apex. (ap, apical cell, cu, cuticle; ep, epidermis; ex, extracellular mucilage; ic, inner cortex; me, medulla; im, intercellular mucilage; oc, outer cortex; sp, spermatangia; spm, spermatangial mother cell; mf, medullary filament; rh, rhizoidal filament; sp, spermatangia; spm, spermatangial mother cell; see, sterile cortical cell). (A) Longitudinal section of gametophyte thallus apex to show many apical cells and their derivative filaments that form the axial filaments. A single periaxial cell is cut off from each axial cell (arrow) and successive peraxial cells rotate in a zig-zag manner along each axial filament. Periaxial cells cut lateral filament that form the cortex. X 1800. (B) Light microscope view of mature thallus to show epidermis, inner and outer cortices and medulla. The extracellular mucilage covering and intracellular mucilage in cortical and medullary regions stain deep violet indicating the presence of both carboxylated and sulphated polysaccharides (TBO). X 1665. (C) Same, magnified to show distribution of polysaccharide secretions. The extracellular and intercellular mucilage in cortical region and cell walls stain deep violet revealing both sulphated and carboxylated polysaccharides. The intercellular mucilage in medullary region stains reddish violet indicating abundance of sulphated over the carboxylated polysaccharides (TBO). X 1800. (D) Longitudinal section of young thalli to show spermatangia scattered over thalli surfaces. The spermatangial mother cell and the tips of the spermatangia are well stained for proteins (arrowhead; CBB). B x 1440. (E) Same, to show autofluorescent compounds present in spermatangial mother cell and spermatangium. X 1440. (F) A young thallus revealing a few sterile cortical cells among the fertile ones. Spermatangia are ovate with darkly staining tips that contain proteinaceous material (CBB) x 1440. (G) Longitudinal section of young thallus to show that the outer cortical cells segment 2 or 3 spermatangial mother cells that produce 1 or 2 spermatangia. The spermatangia mother cells and tips of spermatangia stain well for proteins. X 5625.

REPRODUCTIVE BIOLOGY

SPERMATANGIA: The spermatangia are dispersed superficially on the top of the young branches (Figure 3D). The outer cortical cells cut off 2 to 4 Spermatangial mother cells (Figure 3E,F,G). Each spermatangial mother cell cuts 1 or 2 spermatangia

(Figure 3E,F,G). All the spermatangia are ovoid to ellipsoid in shape and have dark staining tips (Figure 3E,F,G). A few outer cortical cells do not cut spermatangia but produce epidermal cells (Figure 3E,F,G). The extracellular mucilage that veneers the spermatangial mother cells gelatinizes after the formation of spermatangia (Figure 2A) and thus enables the release of spermata.

CARPOGONIAL BRANCH: The carpegonial branch is 3-celled and arises from the inner cortical cells. Occasionally, 2 carpegonial branches arise from a common supporting cell (Figure 4C). The carpegonium is elongate, conical shaped (Figure 4C) and has a long trichogyne (Figure 4C). After fertilization, the carpegonium becomes large and spherical in shape and issues a single connecting filament from its terminal end. **AUXILIARY CELL:** Auxiliary cells are the dedifferentiated inner, spherical Cortical(cells that contain in the core darkly stained nuclei and uninucleate. As Auxiliary cell matures, the juxtaposed, polynucleate, inner cortical cells stain darkly. The auxiliary cell along with these darkly staining cortical cells comprise the auxiliary cell-complex (Figure 4D,E). After diploidization of the auxiliary cell, the neighbouring cortical cells adjoining the auxiliary cell-complex generate elongate, septate, filaments which form the pericarp (Figure 4D,E). A single gonimoblast initial is cut off from the auxiliary cell and later divides transversely to form a closely packed group of gonimoblast cells (Figure 4D,E). As the auxiliary cell enlarges, it produces from its periphery many branched gonimoblast filaments. Interestingly, a few gonimoblast cells combine with the auxiliary cell (Figure 4D,E) which in turn merge, through pit-connection, with adjacent darkly stained supporting (cortical) cells (Figure 4F). The fusion product of all these cells lead to formation of a big fusion cell. Gonimoblast filaments are seen distributed around the the fusion cell (Figure 4F,G). Those branched gonimoblast cells that face towards the pericarp, do not participate in the formation of initial fusion cell but divide and produce the spreading carposporangia (Figure 4F,G). A few of the gonimoblast filaments remain sterile, unbranched, unseptate and connect the fusion cell with the pericarp (Figure 4G). The mature cystocarp shows a large fusion cell in the centre from which branched and unbranched gonimoblast filaments emanate. The terminal or two or three upper cells of these branched gonimoblast filaments mature into carposporangia (Figure 4F,G). Light and Electron microscopic studies show that mature cystocarp is enveloped by three to five layered thick pericarp (Figure 4F) and is embedded in the medullary region. The carpospores are initially elliptical in shape but at release, through the ostiole, become spherical (Figure 4F).

OSTIOLE FORMATION : The cortical cells, adjacent to the cystocarp, proliferate and produce small cells (Figure 4A,B) which raise above the thallus surface (Figure 4A,B). The ostiole is formed by the gelatinization of the pericarp cells followed by dissolution of adjoining cortical cells. The progressive gelatinization of pericarp cells is followed by the dissolution of vegetative cells (Figure 4B). The resultant lysate assembles at the ostiolar region (Figure 4B). The complete gelatinization of extracellular mucilage and degeneration of vegetative cells makes an orifice (Figure 4A,B) above the thallus surface for the carpospores release (Figure 4A,B).

TETRASPORANGIA: Tetrasporangia are scattered all over the thallus surface except at the branch apices and the main axis. Tetrasporangia are cut off laterally by longitudinal divisions of the

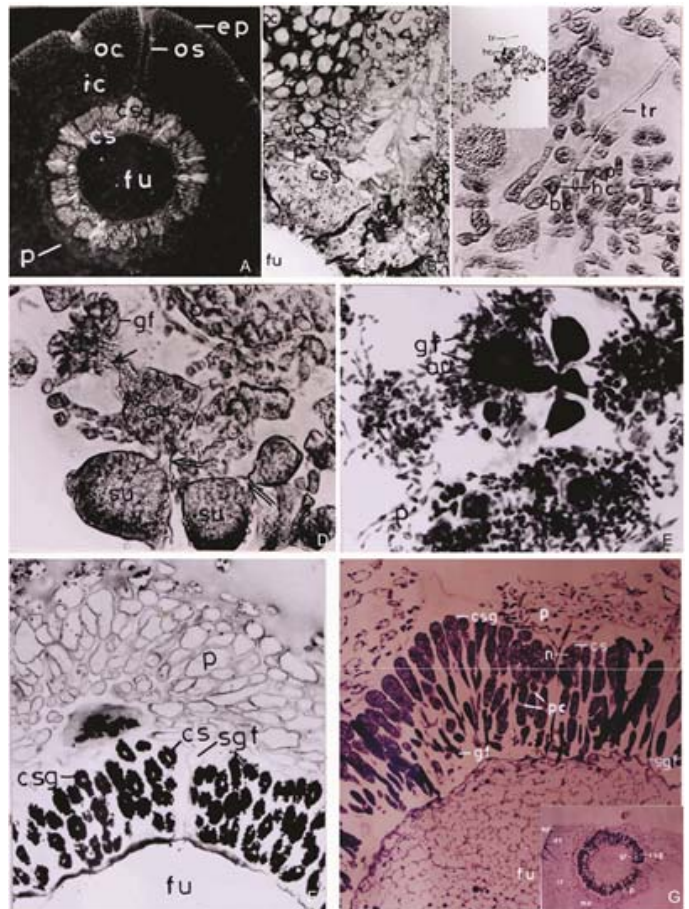


Figure 4. *Solieria robusta*. Carpegonial branch, Auxiliary cell complex, Cystocarp, Fusion Cell, Gonimoblast Filament & Carpospore. (cs, carpospore; csg, carposporangium; ep, epidermis; fu, fusion cell; gf, gonimoblast filament; ic, inner cortex; oc, outer cortex; os, ostiole; me, medulla ; cp, carpegonium; he, hypogynous cell; su, supporting cell; tr, trichogyne; au, auxiliary cell; ; p, pericarp; sgf, sterile gonimoblast filament; cs, carpospore; csg, carposporangium ; n, nucleus; p, pericarp; sgf, sterile gonimoblast filament) (A) Transverse section of mature cystocarp with well developed ostiole. The carposporangia, carpospores, gonimoblast filaments and pericarp cells contain autofluorescent compounds. The epidermal and outer cells are rich in this compound whereas the inner cortical cells show peripheral arrangement. Ax 360; (B) Magnified to show that the pericarp and surrounding vegetative cells lyse. The lysate (arrow) is rich in both sulphated and carboxylated polysaccharides (Photomontage), x 1800 (C) Temporary mounts of thalli to show mature 3-celled carpegonial branches. The hypogynous cell is darkly stained as compared to other two cells of the branch. The carpegonia are conical in shape and trichogynes. In A, two carpegonial branches arise from a common supporting cell, x 1800 (D&E) Gonimoblast initial (arrow) is cut off from the auxiliary cell. Later this initial divides to form a cluster of gonimoblast cells. The auxiliary cell fuses with supporting cells through pit-connections form the fusion cell, x 4500 (F) A portion of the thallus to show the growth of the cortical cells around the cystocarp. Epidermal and outer cortical cells are rich in floridean starch grains as compared to inner cortical and medullary cells, x 1801 (G) Magnified to show that gonimoblast filaments and carposporangia are replete with cellular proteins whereas the fusion cell and cells of the pericarp show depletion of this metabolite, x 2250.

outer cortical cells (Figure 5A,B,C) which later divide transversely to produce one or two additional cells (Figure 5A,B,C). Tetrasporangial initials enlarge in size (Figure 5A,B,C) and

consists thick cytoplasm when equate to the adjacent cortical cells (Figure 5A,B,C). In transactions, mature tetrasporangia show thick and well stained wall (Figure 5A,B,C). A prominent, darkly stained nucleus is apparent in tetrasporangial mother cell (Figure 5E). Immature tetrasporangia are initially oblong in shape but during Progressive maturation elongate basally and expand laterally (Figure 5A,B,C) and appear juxtaposed betwixt the epidermal cells. The cleavage of the protoplast is accompanied by the ingrowth of the septa from the wall of the sporangium (Figure 5A,B,C E). The mature tetrasporangium undergoes zonate, Presumably meiotic, division, to produce four spores of equal size (Figure 5A,B,CE).

HISTOCHEMICAL STUDIES

Axis: The extracellular mucilage that veneers the outer tangential walls of epidermal cells; the walls of epidermis, outer and inner cortical cells and rhizoidal cells stain deep violet with TBO (Figure 3B,C) indicating the presence of carboxylated and sulphated polysaccharides. In *Solieria robusta* the cell walls of the medullary filaments also show identical staining properties. The intercellular mucilage of the cortical cells contain both the above types of polysaccharides whereas that of medullary regions stain reddish violet confirming the abundance of sulphated polysaccharides (Figure 3 C). **Spermatangia:** The extracellular and intercellular mucilages between the epidermal cells and outer cortical cells stain deep-violet indicating the presence of both sulphated and carboxylated polysaccharides. As the cortical cells cut off spermatangial mother cells, which produce spermatangia, the extracellular mucilage gelatinizes and paves way for spermatia release. The spermatangial and spermatial cytoplasm stain moderately and contain sulphated and carboxylated polysaccharides (Figure 3C). The cell walls of spermatangial mother cells and cortical cells stain deep violet indicating the presence of both carboxylated and sulphated polysaccharides. The cell walls of spermatangia bound by polysaccharide covering (Figure 3C). The intercellular mucilage between the spermatangia and spermatangial mother cells is rich in sulphated polysaccharides (Figure 3C). **Cystocarp:** The intercellular spaces and cell walls of fusion cell, gonimoblast filaments and pericarp are rich in both carboxylated and sulphated polysaccharides (Figure 4F). The gonimoblast cell's cytoplasm shows plentiful sulphated polysaccharides as compared to the carposporangial cytoplasm which stains moderately for this metabolite (Figure 4F). The fusion cell and pericarp cells cytoplasm are bereft of this metabolite (Figure 4F). At the ostiolar region, both the pericarp and surrounding cortical cells of the thallus lyse and the lysate of all these cells have copious amount of sulphated and carboxylated Polysaccharides. This process helps in Protection and the dispersal of carpospores. **Tetrasporangia:** The extracellular mucilage, and The cell wall the tetrasporangia stain intensely for wall polysaccharides and reveal carboxylated and sulphated polysaccharides (Figure 5B). The tetrasporangial cytoplasm, however, stains moderately for sulphated polysaccharides (Figure 5B).

LOCALIZATION OF INSOLUBLE POLYSACCHARIDES

Axis: The epidermal and outer cortical cells are replete with floridean starch grains as compared to inner cortical cells (Figure 3F)

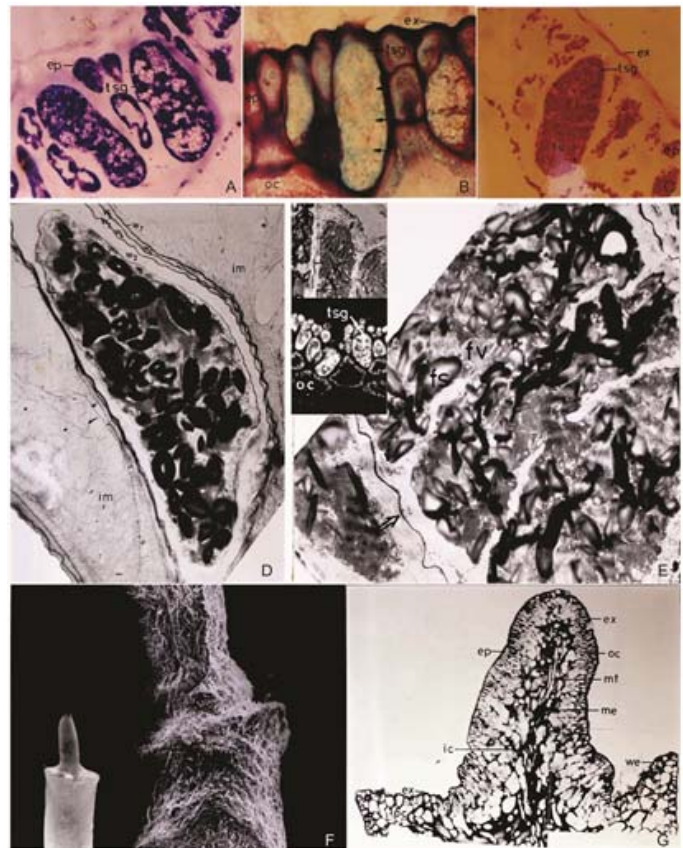


Figure 5. *Solieria, robusta*. Tetrasporangium, Mature carposporangium (ep, epidermis; ex, extracellular mucilage; fs, floridean starch grains; oc, outer cortex; tsg, tetrasporangium; im, intercellular mucilage; ep, epidermis; fs, floridean starch grain; fv, fibrous vesicle; ch, chloroplast; ic, inner cortex; me, medulla; mf, medullary filament; we, wound epidermis) (A). Longitudinal section of the thallus to show mature tetrasporangia. Protoplast cleavage is accompanied by the ingrowth of the septum from the sporangium wall. Proteins are restricted only to cell cytoplasm. Cell walls and intercellular mucilage are bereft of this metabolite (CBB). x 5625. (B). Same, to show in a mature tetrasporangium the extracellular mucilage stains deep violet indicating the presence of sulphated and carboxylated polysaccharides. The tetrasporangium cytoplasm shows metachromasy and confirms the abundance of sulphated polysaccharides (TBO). x 5625 (C). Longitudinal section of thallus to show epidermis veneered by a thick mucilage that stains positively for insoluble polysaccharides. Epidermal cells and outer cortical cells show moderate amount of this metabolite. Tetrasporangium is gorged with floridean starch grains. The tetraspores are zonately arranged (PAS), x 5625 (D). A mature carposporangium showing 7 layered thick cell wall. The layer W1 is electron-dense and thin; W2 is electron-translucent and thick; W3 is electron-dense and thin; W4 is electron-translucent and thick; W5 is electron-dense and thin; W6 is electron-translucent and thick and W7 is electron-dense and thin. This is followed by intercellular mucilage with reticulate texture, x 8160 (E). Transmission electron micrograph to show tetrasporangium. The cleavage furrows are seen as invaginations of the plasmalemma. These furrows show centripetal extension (arrows) and partition the tetrasporangium into four zonately arranged tetraspores. x 10,600. L. S. of thalli showing autofluorescent compounds in tetrasporangium. (F). Scanning electron micrograph to show regenerate growth (arrow) from the tip of a wound tissue, x 224. Blade-like outgrowths (arrows) regenerate from wound portions of the plants, x 10 (G). Longitudinal section of a regenerating young branch from the wound surface. The epidermal layer covering the new tissue is easily distinguished from that of the wound surface. This indicates

regeneration of cells from the wound surface of the medullary region. Both the original and the regenerated tissues are covered by thick mucilage that stains for both sulphated and carboxylated polysaccharides. The intercellular mucilage both in apical and cortical region reveals the presence of carboxylated and sulphated polysaccharides, x 1800.

Spermatangia : The cell wall of spermatangia and spermatangial mother cells stain feebly for wall polysaccharides. **Cystocarp :** The cell walls of fusion cell, gonimoblast filaments and pericarp are rich in insoluble polysaccharides but that of carposporangia stain with less intensity (Figure 4F). **Tetrasporangium :** The extracellular mucilage that covers both the outer tangential walls of the epidermis and tetrasporangium is replete with polysaccharide secretions. The walls of epidermal cells and tetrasporangia stain well for insoluble polysaccharides (Figure 5C). The tetrasporangium and tetraspores are gorged with floridean starch grains (Figure 5C).

LOCALIZATION OF TOTAL PROTEINS

Axis : The cytoplasmic proteins in the epidermal cells and the outer cortical cells show uniform distribution, whereas in the inner cortical cells they are polarized and restricted to cell periphery (Figure 3D E ,F ,G). In apical region the cytoplasm of apical cell and their derivative cells (axial filament) is rich cytoplasmic proteins. **Spermatangia :** Spermatangial mother cells and outer cortical cells are rich in cytoplasmic proteins (Figure 3F,G). The Spermatangia are polarized with dark staining, protein-rich, tips (Figure 3F, G). The lower-half of the spermatangium is, however, protein negative (Figure 3F,G). **Cystocarp :** The mature cystocarp shows differential distribution of cytoplasmic proteins (Figure 4G). The cytoplasm of gonimoblast filaments is rich in this metabolite as compared to that of the carposporangia (Figure 4G). The carposporangia and the carpospores are uninucleate and contain abundant cytoplasmic proteins (Figure 4G) whereas the fusion cell and the pericarp cell show a low ebb of this metabolite (Figure 4G). **Tetrasporangia :** The mature tetrasporangia and the tetraspores are replete with cytoplasmic proteins (Figure 5A).

LOCALIZATION OF NUCLEIC ACID

In the cytoplasm of carposporangia and carpospores, nuclei and extracellular materials stain well with aceto-iron- haematoxylin and chloral hydrate.

LOCALIZATION OF AUTOFLUORESCENT COMPOUNDS

The thallus sections when observed under UV light (blue-violet) reveal many auto fluorescent compounds that are localized in both epidermal and cortical cells. The epidermal cells are fully gorged with auto fluorescent compounds that are uniformly distributed in cell cytoplasm (Figure 3E). In the inner cortical cells, auto fluorescent compounds occur in peripheral cytoplasm or they lie close to the cell wall (Figure 3E). The tips of spermatangia and spermatangial mother cells (Figure 3E) are rich in auto fluorescent compounds. In mature cystocarp, auto fluorescent compounds are localized in carpospores, carposporangia and in gonimoblast filaments (Figure 4A). The gonimoblast filaments are rich in this compound as compared to the carpospores. The cell walls of pericarp and fusion cells show very weak fluorescence (Figure 4A).

The tetrasporangium and tetraspores are replete with auto fluorescent compounds (Figure 5E).

ULTRASTRUCTURAL STUDIES OF GAMETOPHYTE AND TETRASPOROPHYTE THALLI

Epidermis : The epidermal cells are embedded in an amorphous matrix and cell walls show reticulate arrangement of microfibrils. **Outer Cortex :** The outer cortical cells show multilayered fibrillar cell wall. The cytoplasm is replete with floridean starch grains. **Inner Cortex :** The inner cortical cells show less floridean starch grains (Figure 2D) as compared to epidermal and outer cortical cells. In tetrasporic plants both disc and elliptical shaped chloroplasts coexist. The mitochondria show tubular cristae which in cross section appear circular. **REPRODUCTIVE STRUCTURES:** Carposporangia: The young carposporangial cytoplasm has dictyosomes, chloroplasts and a few floridean starch grains (Figure 5D). The dictyosomes produce vesicles. The young carposporangium is enclosed in mucilaginous sheath. The mature carposporangium has multilayered cell wall and at its base possesses a large vacuole that is filled with fibrous materials. These layers show parallel arrangement of microfibrils (Figure 5D). **Tetrasporangia:** A thick and electron-transparent, mucilaginous sheath produced through the release of fibrous contents of vesicles, encloses the tetrasporangial cytoplasm. The tetrasporangial wall is composed of electron-dense, microfibrils and is distinguished from epidermal cell wall which possesses Parallel arrangement of microfibrils (Figure 5E). The mature tetrasporangium is gorged with floridean starch grains (Figure 5E) as compared the adjacent epidermal cells which show little storage materials (Figure 5E). Mature tetrasporangial cytoplasm show fibrous Vesicles which fuse directly with plasmalemma and release their contents or they fuse with one another to form large, fibrous, vacuoles (Figure 5E). The chloroplasts are also seen in the cell cytoplasm. The protoplast cleavage is accompanied by the cleavage furrows from the plasmalemma (Figure 5E). These cleavage furrows show centripetal extension and partition the tetrasporangium into four zonately arranged tetraspores. The tetrasporangium undergoes simultaneous zonate division producing four spores. The initial median cleavage is arrested just short of completion, and other two cleavages are initiated at the same depth (Figure 5E).

WOUND REGENERATION

Tissue damaged is caused either by wave action or by heavy epiphytic growth. The cells of the wound surface become mitotically active and form the epidermal layer. After the formation of epidermis the medullary regions cells retain their mitotic activity and develop into the regenerated branch(Figure 5F,G). The anatomy of the regenerated branch is identical to that of the normal branch. Both the normal and the regenerated tissues have thick extracellular and intercellular mucilages that stain for both sulphated and carboxylated polysaccharides (Figure 5G).

DISCUSSION

The plants of *Solieria robusta*, from Port Okha, grow luxuriantly attached to the calcareous rocks in rock-pools of the intertidal regions either through discoid or hapteroid holdfasts. The red algae serve as important source for various phytochemicals, health

beneficial extracts⁵⁻⁸ and number of other uses. The learning of fundamental information about algae⁸⁻¹¹ should be considered one important aspect to generate the value to rich information of different species. The constituent of algae depends upon the type of algae and life cycle to respective species.

Generally, the Solieriacean plants develop discoid holdfasts when they are attached to mussel-shells or small stones. According to Chapman (1973),¹² the macroscopic algae (seaweeds) that grow in the intertidal and subtidal regions are always attached to the substratum by discoid holdfasts which are either cellular being extension of basal cell; prostrate and filamentous; or discoid. Perrone and Cecere (1994)¹³ separate *Agardhiella subulata* and *Solieria filiformis* on the basis of holdfast morphology, the former being attached by means of simple discoid holdfast, whereas the latter grows erect from the fibrous basal system. My work on *Solieria robusta* agrees with that of Chapman (1973)¹² that intertidal algae are attached through discoid holdfasts. *Solieria robusta* supports abundant epiphytic algal growth (Present work) as seen in many marine seaweeds (Sieburth and Tootle, 1981).¹⁴ *Gastroclonium Iyengarii* supports epiphytic bacterial growth on its thallus which is responsible for iridescence. According to Duckett and Knox (1984)¹⁵ epiphytism is a common process in marine plants particularly in a biological communities where substratum is the limiting factor. In *Solieria robusta* growth is started by a group of apical cells where the apical cell divides obliquely producing subterminal cells which cut off periaxial cells. Successive periaxial cells rotate around the axial filament and later divide to produce branched cortical filaments (Present work). Similar type of apical development is observed in *S. Chordalis* and *S. Tenera* and also in many other uniaxial taxa like *Rhabdonia*, *Areschougla* and *Melanema*.¹⁶ In *S. robusta* the anatomical differentiation of the thallus into epidermis, outer and inner cortices and medulla with their associated polarized distribution of floridean starch grains and cytoplasmic proteins is a remarkable adaptation which confers on the plant both ecological and physiological survival strategies to withstand tidal fluctuations and grow luxuriantly in the intertidal regions (Present work). The scanning electron microscopic observations on floridean starch grains of *Seirospora griffithsiana* show Morphological variations of starch grains.¹⁷ Boney (1975, 1978)¹⁸⁻²⁰ observes large range of starch grains size in carpospores of *Bonnemaisonia nootkana* as well as in carpospores and holdfast of *Rhodymenia pertusa*. Meeuse et al. (1960)²¹ have observed wide range of diameter in starch granules in *Odonthalia floccosa*. Floridean starch grains are variable in shape and size due to their scattered disposition in the cytoplasm where they are constantly subjected to stress conditions. In *Solieria robusta* transmission electron microscopic studies show that in the inner cortical cells the major organelles like chloroplasts and mitochondria are present in the peripheral region and lie close to the cell wall. In *Solieria robusta* a thick extracellular mucilage covering consisting of a mixture of carboxylated and sulphated polysaccharides veneers the epidermis surface (Present work). These extracellular substances are composed mainly of mucopolysaccharides.^{22,23} The cuticle protects the algae from desiccation and predation.²⁴ My work supports this contention. In *Solieria robusta* the intercellular mucilage is fibrous in nature and is replete with sulphated and carboxylated polysaccharides (Present work). My work concludes

that intercellular mucilage provides strength as well as flexibility to the thallus and protects the seaweeds against physical damage and constant environmental stress conditions. In *Solieria robusta* both epidermal, cortical and medullary cell walls stain deeply for carboxylated and sulphated polysaccharides (Present work). The walls of *Nizyenia australis* and *Scinaia forcellata* and *Asparagopsis taxiformis* are composed mainly of carboxylated and sulphated polysaccharides.² The cell walls of *Sargassum vulgare* and *S. johnstonii* are also made up of carboxylated and sulphated polysaccharides.²⁵ The presence of polysaccharides in cell walls of a number of brown algae has been demonstrated by X-ray diffraction²⁶ and differential staining techniques.^{27,28} Frei and Preston (1964)²⁹ suggest that these polysaccharides, in addition to ion exchange, play a strong structural role. The walls of red algal cell typically have a layered fibrillar appearance enmeshed in a mucopolysaccharide matrix. In *S. robusta* the cell wall is a multilayered, fibrillar structure embedded in amorphous matrix. In *Solieria robusta* spermatangia occur in large patches on the surface of young branches (Present work). Yamamoto (1975)³⁰ recognized 5 types of spermatangial developmental patterns. (a) *Verrucosa* type, (b) *Textorii* type, (c) *Chorda* type, (d) *Symmetrica* type, (e) *Henriguesiana* type. In *Solieria robusta* (Present work) the spermatangial mother cells develop from the cells of outer cortex. A few cortical cells remain sterile and do not cut off spermatangia and thus the spermatangia do not form a continuous layer over the thallus surface. The development of the spermatangia, thus, conforms to the *Symmetrica* type. According to Min-Thein and Womersley (1976)³¹ in *Solieria robusta* the cortical cells cuts off 3 or 4 spermatangial mother cells, each of which produces 2 or 3 ovoid spermatangia. Gabrielson and Hommersand (1982),¹⁶ in *Solieria tenera*, observed surface cortical cells produce 2 to 5 spermatangial mother cells, which in turn produce successively spermatangia. My work supports the view of Min-Thein and Womersley (1976)¹⁶ regarding the development of spermatangia. In *Scinaia forcellata* (Vijayaraghavan and Bhatia (1996)²² and *Solieria robusta* (Present work) histochemical studies reveal that spermatangial cytoplasm contain mucopolysaccharides and probably contribute to the mucilage layer which covers the spermatangia) It is possible that these mucilages exert constant pressure on spermatangial wall and thus play an important role in spermatia release. The carpogonial branch of *Solieria robusta* is 3-celled and consists of carpogonium, hypogynous and basal cells. Min-Thein and Womersley (1976)¹⁶ in *S. robusta* and Gabrielson and Hommersand (1982)³² in *S. tenera* and *S. chordalis* and *Agardhiella subulata* observed identical carpogonial branches. In *Sarconema* sp. the carpogonial branch is either 3 or 4 celled.³³ Controversy exists whether *Solieria* is procarpic or nonprocarpic. Kylin (1932)³⁴ in *S. chordalis* and Min-Thein and Womersley (1976)¹⁶ in *S. robusta* from southern Australia report them to be nonprocarpic whereas Wynne and Taylor (1973)³⁵ in *S. chordalis* observed procarpic nature. My work on *S. robusta* from Port Okha confirms it to be nonprocarpic. The genera *Agardhiella* and *Sarcoditheca* also possess an auxiliary cell-complex that differentiates prior to diploidization of the auxiliary cell. *Sarconema filiformis* and *S. scinaoides* and *Placentophora* sp.,³⁶ however, have undifferentiated auxiliary cells prior to diploidization. Kylin (1932)³⁴ observed that the auxiliary cells in

Solieria are not recognizable before fertilization. Contrary to this statement, in *Solieria robusta*, the auxiliary cell is prominent and can be distinguished from the cortical cells before diploidization (Present work). It appears that the dedifferentiation of the inner cortical cell to form the auxiliary cell confers on it special attributes where this cell differs both morphologically and functionally and produces only gonimoblast initials. In *Solieria robusta* (Present work) after diploidization of the auxiliary cell, the adjacent cortical cells form branched chains of cells which envelope the auxiliary cell-complex and auxiliary cell. A few inner gonimoblast filaments and adjacent darkly stained cells fuse with auxiliary cell and their fusion Product leads to the formation of large fusion cell with a common boundary. In early stages, fusion cell shows lysis of cell walls in the centre of matrix (Present work). Kylin (1932)³⁴ in *Solieria chordalis* and Min-Thien Womersley (1976)³¹ in *S. robusta* suggest that fusion cell is formed by the coalescence of the first formed gonimoblast cells with the auxiliary cell. Gabrielson and Hommersand (1982)¹⁶ in *S. chordalis* and *S. tenera* report that fusion cells are formed by the fusion of auxiliary cell, gonimoblast cells and cells of the lateral vegetative filaments proximal to the auxiliary cell. My work on *Solieria robusta*, from the Port Okha, reveals that the fusion cell is formed by auxiliary cell, cells of the auxiliary cell- complex and a few inner gonimoblast cells and supports the contention of Gabrielson and Hommersand (1982).¹⁶ More ultrastructural and histochemical studies are needed to understand the structure, nature and function of this enigmatic cell-complex in different orders of Rhodophyceae. According to Kugren and West (1973)³⁷ in *Levringiella gardneri* (parasitic taxon), the cells of the pericarp are secretory in nature and produce mucilage that acts as bacteriostatic agent and protects the carpospores from desiccation. In *Solieria robusta*, from Port Okha, ultrastructural and histochemical studies reveal that the cell wall of pericarp is made up of electron-dense, fibrillar materials and is rich in complex Polysaccharides. According to Min-Thien and Womersley (1976),³¹ in *Solieria robusta*, the terminal carposporangia arise from the gonimoblast filaments. In *S. tenera* and *S. chordalis* only the terminal cells of the branched gonimoblast becomes carposporangia. In contrast, *Solieria robusta*, from Port Okha, reveals that both terminal and two or three upper cells of the gonimoblast filament differentiate into carposporangia. Gabrielson and Hommersand³² in *Agardhiella subulata* reported both terminal as well as carposporangia in succession. Papenfuss and Edelstein (1974)³³ observed chains of carposporangia from the tips of gonimoblasts of *Sarconema scinaoides*. The carposporangia development is variable in different taxa and need not be used as taxonomic character. My work on *Solieria robusta* supports the views of Scott and Dixon (1973)³⁸ and reveals that fibrous vacuole at the base of the carpospores helps in the dispersal of carpospores). In Florideophycideae two distinct types of zonately divided tetrasporangia occur. The first and most common is the successive type where median division occurs initially and is followed by two other divisions. In the second, simultaneous type, all the cleavages are simultaneously initiated. In *Solieria robusta* the ultrastructural studies reveal that the median cleavage is arrested just short of completion and other cleavages are later initiated (Present work). Guiry (1990)³⁹ found that in *Agardhiella subulata* (Solieriaceae, Gigartinales), after the first meiotic division, the median cleavage

furrow is almost complete but stops just short of merging. Following the second meiotic division, the other two furrows invaginate and once they come at the same depth as the first cleavage furrow, all three simultaneously merge. Guiry (1990)³⁹ called this as intermediate between successive and simultaneous zonate division. In *Solieria robusta*,^{2,3,40} mature tetrasporangial cytoplasm shows the presence of chloroplasts, fibrous vesicles and abundant floridean starch grains. The starch grains are variable in shape and size. Kugrens and West (1972)³⁷ observed that thickening of tetraspore wall prevails as the tetraspore mother cell divides and a dark staining fibrillar structural shape outer wall layer in the middle of tetraspores and vegetative cell walls, causing the points of weakness. The Inner layer is electron-transparent and is identical with that of fibrous vesicles. It clearly shows that these vesicles release mucilages that form the mucilaginous layer or sheath which encloses the tetrasporangial cytoplasm and further help in attachment of tetraspore to the substratum.

Wound Regeneration: The process of regeneration at the sub cellular level has been studied in *Sargasson muticum*. The mitosis is preceded by increase in cytoplasmic density and cell organelle numbers suggesting increased metabolic activity. Similar results have been observed in other algae species.⁴¹ In solieria filiformis regeneration showed marked polarity by progressing differently at the proximal and distan ends of an explanat.¹³ *Solieria robusta* also show the process of regeneration.

SUMMARY

The plants of *Solieria robusta* are collected from Port Okha (Gujarat). The Okha-reef comprises of limestone rocks that are occasionally covered with calcareous deposits. (*S. robusta* grows luxuriantly and is attached to the limestone rocks, in the rock pools, of intertidal region. A few plants are occasionally washed ashore as drifts. Both gametophyte and tetrasporophyte plants are attached to the rocks through discoid holdfasts or hapteron-like structures. The thalli are purplish-red to brownish-red in colour and consist of terete or slightly compressed, erect lateral branches that are attenuate at the base and subacute at the tip. Branching is irregular and occasionally from the' damaged portion 4 or 5 branches regenerate to form an umbel. The thallus is multiaxial and at the apex has a group of apical cells which initially divide obliquely to produce subterminal cells which are the progenitor of periaxial cells that segment laterally cutting filaments that divide to form the cortex. The central derivatives of the apical cell becomes the axial filament. The medulla is composed of axial filament, interconnecting rhizoidal filaments. The thallus is, thus, differentiated into cortex and medulla. The cortical region comprises the epidermis, outer and inner cortices. In male plants, the outer cortical cells produce 2 to 4 spermatangial mother cells. Spermatangia are ovoid to ellipsoid in shape and possess dark staining tips. The 3-celled carpogonial branch, arises from the inner cortical cell and consists of carpogonium with long trichogyne, hypogynous cell and basal cell. Auxiliary cell and the cells of the auxiliary cell-complex, stain darkly prior to diploidization and can be easily recognized from other cortical cells. It is a nonprocarpic plant where connecting filament arises from the fertilized. As the auxiliary cell matures additional gonimoblast filaments arise from its periphery. The auxiliary cell, along with a few basal gonimoblast

cells and darkly stained adjacent cortical cells fuse and the fusion product of all these cells lead to the formation of large, round, fusion cell in the centre of the mature cystocarp. The terminal one or two cells of each branched gonimoblast filaments becomes the carposporangia. A few gonimoblast filaments remain sterile, become nonseptate and unbranched and connect the pericarp with the fusion cell. The carpospores are released through a ostiole that is formed by the gelatinization of pericarp and surrounding vegetative cells. Tetrasporangia are scattered all over the thallus surface except the branch apices and the main axis. The tetrasporangial initials enlarge, undergo meiotic divisions, and produce four, equal, zonate tetraspores. The mucilage that veneers the epidermis, cortical, rhizoidal and medullary cell walls stains deep violet indicating the presence of both carboxylated and sulphated polysaccharides. In cortical region, the intercellular mucilage shows both sulphated and carboxylated polysaccharides. The epidermal and outer cortical cells are rich in floridean starch grains and cytoplasmic proteins as compared to the inner cortical and medullary cells. The spermatangial mother cell walls and spermatangium are rich in carboxylated and sulphated polysaccharides. Spermatangial mother cell shows presence floridean starch grains and proteinaceous materials. In the spermatangium the protein shows polarized distribution. The carposporangia and carpospores are replete with floridean starch grains and sulphated polysaccharides. The cytoplasm of gonimoblast filaments and carposporangia show plenty of proteins. Tetrasporangia and tetraspores stain well for wall polysaccharides whereas their cytoplasm is replete with floridean starch grains and proteins. Tetrasporangia and tetraspores stain well for wall polysaccharides whereas their cytoplasm is replete with floridean starch grains and proteins. Carposporangial cytoplasm is fully packed with floridean starch grains, fibrous vesicles, and chloroplasts. Fibrous vesicles fuse with plasmalemma and participate directly in the cell wall formation. Carposporangial wall is 3 to 5 layered. Tetrasporangia are fully packed with floridean starch grains as compared to the contiguous epidermal cells. The tetrasporangial cytoplasm has fibrous vesicles and chloroplasts. These vesicles either fuse directly with the plasmalemma or fuse among themselves to form large vacuoles. The development of zonate tetraspores is simultaneous.

CONCLUSIONS

The conclusive points are: The thallus is attached to the limestone rocks by a discoid holdfast or the hapteron-like structure. Branching is irregular, occasionally, 4 or 5 branches arise from a damage branch to form an umbel. Epidermis is veneered by a thick extracellular mucilage which is rich in both carboxylated and sulphated polysaccharides. Multiaxial thallus growth is triggered by a group of apical cells. The thallus consists of cortex and medulla. The cortex is further differentiated into epidermis, outer and inner cortices. Spermatangia are disseminated on the surface of young branches. Outer cortical cells produce 2 to 4 spermatangial mother cells which in turn form one or two spermatangia. The spermatangia are ovoid to ellipsoid in shape and polarized with dark staining protein-rich tips. The extracellular mucilage covers the thallus surface and gelatinizes only after the formation of spermatangia. This process enables the release of spermatia. The

carpogonial branch is 3-celled and arises from the inner cortical cells. Any enlarged inner cortical cell, with darkly staining nucleus, acts as an auxiliary cell. The fusion product of auxiliary cell, inner gonimoblast cells and adjacent darkly stained cortical cells lead to the formation of large, round, fusion cell with a common boundary. The carpospores are escaped through the ostiole which is formed by the lyses of pericarp and vegetative cells. The lysate is rich in sulphated and carboxylated polysaccharides. Autofluorescent proteinaceous materials are present in epidermal and cortical cells. Spermatangia carposporangia, gonimoblast filaments and tetrasporangia also possess such autofluorescent materials. Tetrasporangia are scattered all over the thallus surface. The tetrasporangium cytoplasm is gorged with floridean starch grains and proteins and show fibrous vesicles and chloroplasts. The tetraspores are zonately arranged. *Solieria robusta* (Greville) Kylin shows regeneration without callus formation.

ACKNOWLEDGEMENTS

Authors acknowledge the cooperation of the Department of Botany, University of Delhi. All the research work was carried out in the laboratory of the department in the guidance of Professor M.R. Vijayaraghavan.

REFERENCES AND NOTES

1. I. Nashier Gahlawat, P. Lakra, J. Singh, B.S. Chhikara. Developmental and histochemical studies on carposporophyte of *Solieria robusta* (Greville) Kylin (Solieriaceae, Gigartinales) from Port Okha, India. *J. Integr. Sci. Technol.* **2020**, 8 (2), 12–20.
2. I. Nashier, A. Jha. Developmental and histochemical studies on Spermatangia in *Solieria robusta* (Greville) Kylin (Solieriaceae), Rhodophyta. In *Plant Form and Function*; Bhatia, B., Shukla, A. K., Sharma, H. L., Eds.; Angkor Publishers, **1998**.
3. I. Nashier, A. Jha, M.R. Vijayaraghavan. Tetraspore development in *Solieria robusta* (Greville) Kylin (Solieriaceae, Gigartinales) from Port Okha, India. *Phytomorphology* **1999**, 49 (3), 233–240.
4. M.R. Vijayaraghavan, I. Nashier, B. Mam. In vitro studies of *Callithamnion* sp. (Ceramiaceae, Ceramiales): Growth and differentiation. *Seaweed Res. Utiln.* **1995**, 17, 13–22.
5. P. Lakra, I. Nashier Gahlawat. Prospective Phytochemicals for alleviation of different chronic ailments. *Integr. J. Soc. Sci.* **2015**, 2 (1), 36–39.
6. P. Lakra, I. Nashier Gahlawat. The role of Nutrition in the Immune system functions. *Integr. J. Soc. Sci.* **2016**, 3 (1), 30–33.
7. I. Nashier Gahlawat, P. Lakra. Contextual implicit role of PROBIOTICS in improving the Human Health. *J. Integr. Sci. Technol.* **2017**, 5 (2), 50–53.
8. P. Lakra, S. Sehgal, I. Nashier Gahlawat, M. Wadhwa nee Dasbas. Use of products developed from potato flour, defatted soy flour and corn flour in combating malnutrition. *Integr. J. Soc. Sci.* **2018**, 5 (1), 47–50.
9. M. Wadhwa, I. Nashier Gahlawat, P. Lakra, S. Nischal. Children's Questions in Science Classrooms: A Potential Source of Learning. *Integr. J. Soc. Sci.* **2018**, 5 (2), 41–46.
10. M. Wadhwa, I. Nashier Gahlawat, A. Chhikara. Understanding Students' Alternative Conceptions: A Mirror to their Thinking. *Integr. J. Soc. Sci.* **2020**, 7 (1), 9–13.
11. M. Wadhwa nee Dabas, K. Kaur. Child's Construction of Knowledge: Role of Activities in Classroom. *Integr. J. Soc. Sci.* **2017**, 4 (1), 20–25.
12. D.J. Chapman. *The algae*; Springer, **1973**.
13. C. Perrone, E. Cecere. Two Solieriacean Algae New To the Mediterranean:

- Agardhiella Subulata and Solieria Filiformis (Rhodophyta, Gigartinales). *J. Phycol.* **1994**, 30 (1), 98–108.
14. J.M.N. Sieburth, J.L. Tootle. Seasonality of Microbial Fouling on *Ascophyllum Nodosum* (L.) Lejol., *Fucus Vesiculosus* L., *Polysiphonia Lanosa* (L.) Tandy and *Chondrus Crispus* Stackh. *J. Phycol.* **1981**, 17 (1), 57–64.
 15. S.C. Ducker, R.B. Knox. Epiphytism at the Cellular Level with Special Reference to Algal Epiphytes. *Cell. Interact.* **1984**, 113–133.
 16. Gabrielson P. W., Hommersand M. H. The Atlantic Species of *Solieria* (Gigartinales Rhodophyta): Their Morphology Distribution and Affinities. *J. Phycol.* **1982**.
 17. R.G. Sheath, J.A. Hellebust, T. Sawa. Ultrastructure of the floridean starch granule. *Phycologia* **1981**, 20 (3), 292–297.
 18. A.D. Boney. The liberation and dispersal of carpospores of the red alga *Rhodymenia pertusa* (Postels et Rupr.) J. Ag. *J. Exp. Mar. Bio. Ecol.* **1978**, 32 (1), 1–6.
 19. A.D. Boney. Mucilage sheaths of spores of red algae. *J. Mar. Biol. Assoc. United Kingdom* **1975**, 55 (3), 511–518.
 20. A.D. Boney. Survival and growth of alpha-spores of *Porphyra schizophylla* Hollenberg (Rhodophyta: Bangiophyceae). *J. Exp. Mar. Bio. Ecol.* **1978**, 35 (1), 7–29.
 21. B.J.D. Meeuse, M. Andries, J.A. Wood. Floridean starch. *J. Exp. Bot.* **1960**, 11 (2), 129–140.
 22. M.R. Vijayaraghavan, B. Bhatia. Developmental and histochemical studies on the spermatangium and carposporophyte of *Scinaia forcillata* (Nemaliales, Rhodophyta) from Port Okha, India. *Nov. Hedwigia* **1996**, 112, 135–146.
 23. M. Vijayaraghavan. Developmental and histochemical studies on *Nizymania australis* Sonder (Nizymaniaceae, Gigartinales, Rhodophyta). *Proc. Indian Natl. Sci. Acad. Part B Biol. Sci.* **1991**, 57 (1), 69–76.
 24. W.H. Gerwick, N.J. Lang. Structural, Chemical and Ecological Studies on Iridescence in *Iridaea* (Rhodophyta). *J. Phycol.* **1977**, 13 (2), 121–127.
 25. M. Vijayaraghavan, I. Kaur. Ultrastructure and histochemistry of vegetative Thallus in *Sargassum vulgare* C. Agardh and *S. johnstonii* Setchell & Gardner. *Proc. Indian Natl. Sci. Acad. Part B Biol. Sci.* **1991**, 57 (5), 319–328.
 26. E. Frei, R.D. Preston. Configuration of alginic acid in marine brown algae. *Nature* **1962**, 196 (4850), 130–134.
 27. L. V. Evans, M.S. Holligan. Correlated Light and Electron Microscope Studies on Brown Algae I. Localization of Alginic Acid and Sulphated Polysaccharides in Dictyota. *New Phytol.* **1972**, 71 (6), 1161–1172.
 28. M.M. E. Histological studies on the genus *Fucus*. I. Light microscopy of the mature vegetative plant. *Protoplasma* **1966**, 62, 287.
 29. E. FREI, R.D. PRESTON. Non-Cellulosic Structural Polysaccharides in Algal Cell Walls I. Xylan in Siphonous Green Algae. *Proc. R. Soc. Lond. B. Biol. Sci.* **1964**, 160, 293–313.
 30. H. Yamamoto. The Relationship between *Gracilariopsis* and *Gracilaria* from Japan. *Bull. Fac. Fish. Hollaido Univ.* **1975**, 26 (3), 217–222.
 31. U. Min-Thein, H.B.S. Womersley. Studies on southern Australian taxa of Solieriaceae, Rhodoniaceae and Rhodophyllidaceae (Rhodophyta). *Aust. J. Bot.* **1976**, 24 (1), 1–166.
 32. P.W. Gabrielson, M.H. Hommersand. the Morphology of *Agardhiella Subulata* Representing the Agardhielleae, a New Tribe in the Solieriaceae (Gigartinales, Rhodophyta). *J. Phycol.* **1982**, 18 (1), 46–58.
 33. G.F. Papenfuss, T. Edelstein. The morphology and taxonomy of the red alga *Sarcocnema* (Gigartinales: Solieriaceae). *Phycologia* **1974**, 13 (1), 31–43.
 34. H. Kylin. Die Florideenordnung Gigartinales. *Lunds Univ. Arsskr. NF Avd. 2* **1932**, 28, 1–88.
 35. M.J. Wynne, W.R. Taylor. The status of *Agardhiella tenera* and *Agardhiella baileyi* (Rhodophyta, Gigartinales). *Hydrobiologia* **1973**, 43 (1–2), 93–107.
 36. G.T. Kraft. Morphology of *Placentophora* (Solieriaceae, Gigartinales: Rhodophyta), a New Genus Based on *Sarcodiotheca Colensoi* From New Zealand. *J. Phycol.* **1975**, 11 (4), 399–410.
 37. P. Kugrens, J.A. West. the Ultrastructure of Carposporogenesis in the Marine Hemiparasitic Red Alga *Erythrocytis Saccata*. *J. Phycol.* **1974**, 10 (2), 139–147.
 38. J.L. Scott, P.S. Dixon. Ultrastructure of Tetrasporogenesis in the Marine Red Alga *Ptilota Hypnoides*. *J. Phycol.* **1973**, 9 (1), 29–46.
 39. M.D. Guiry. Sporangia and spores. In *Biology of the red algae*; **1990**; pp 347–376.
 40. I. Nashier, A. Jha, M.R. Vijayaraghavan. Wound regeneration in *Solieria robusta* (Greville) Kylin (Solieriaceae, Gigartinales) from Port Okha, India. *Seaweed Res. Utiln.* **1998**, 20 (1&2), 119–124.
 41. J. McLachlan, L.-M. Chen. Formation of adventive embryos from rhizoidal filaments in sporelings of four species of *Fucus* (Phaeophyceae). *Can. J. Bot.* **1972**, 50 (9), 1841–1844.