Fig. 6. Cleavage would then occur at points spaced multiples of exactly 10.0 bases apart. In the case of a nucleosome, in contrast, with only two superhelical turns, the lower surface of the lower turn and the upper surface of the upper turn are also exposed to enzyme digestion, and attack should occur on the average at angles of θ_1 and θ_2 from the perpendicular, as is indicated by solid arrows in Fig. 6. If $\theta_1 + \theta_2$ were 108°, or three times the angle between adjacent base pairs, then fragments produced by cuts one turn of the superhelix apart would be increased from 80 to 83 bases. For $\theta_1 + \theta_2 = 144°$, the fragment length would become 84 bases. The lengths are increased rather than decreased compared to an exact multiple of 10.0 bases because the double helix is right-handed, while the superhelix is left-handed. This effect can be seen not only in Fig. 6, but also in a detailed drawing of a left-handed superhelix used to explain the relative frequencies of cutting by DNase I at various sites in the nucleosome (*9*). The arguments concerning fragment lengths and frequencies of cutting are of essentially the same type, invoking steric bias in the ease of access by an enzyme to the two portions of the same strand of DNA in the two turns of a nucleosome. In summary, the periodicity of digestion of DNA coiled in a two-turn nucleosome is expected to be greater than the periodicity of the double helix itself. Specifically, fragments that are multiples of about 10.4 bases could be produced from DNA with a helical repeat of 10.4 base pairs.

The distribution of fragment lengths determined in our work does not reflect the actual location of cleavage sites but only the distances between them. In further studies, the locations of cleavage sites within the core particle of the nucleosome have been established (*7*). The sites are spaced at multiples of about 10.4 bases, but the precise spacing is variable. The consistence of these further data with the results we report here has been checked by using the locations of sites along with the probability of cleavage at each site (*9*) to calculate a distribution of fragment lengths. This calculated distribution is in reasonable agreement with the measured distributions displayed in Fig. 5. The variation in the spacing of cleavage sites and in the periodicity of fragment lengths may reflect variation in the helical repeat of DNA in the nucleosome. Alternatively, the helical repeat may be constant while the angle of attack by the enzyme varies from one cleavage site to the next (rather than changing in a uniform manner as might be inferred from Fig. 6). The exact interpretation of any digestion data will be rather speculative until much more is learned about nucleosome structure.

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18. Supported by NIH grant CA 20768 to R.D.K., and by an NIH postdoctoral fellowship to L.L. We thank F. Fuller for plasmid pOP203-1, M. Calos for plasmid pMC1, and M. Gait for the dpTTCTGTTGA.

17 August 1978; revised 5 March 1979

"Transfer Connections": Specialized Pathways for Nutrient Translocation in a Red Alga?

Abstract. *"Transfer connections" are morphologically and developmentally distinct pit connections in* Polysiphonia *(Ceramiales). They are intracellular rather than extracellular and have been observed between all cells of the diploid carposporophyte plus those specialized cells of the gametophyte suspected of providing nutritive materials to it.*

Cytoplasmic channels between animal cells (for example, gap junctions) and between plant cells (for example, plasmodesmata) permit intercellular transport and communication which appear necessary for differentiation (*1*). In the complex tissues of some green and brown algae, plasmodesmata are likewise believed to permit transport (*3*). However, plasmodesmata per se are absent in the red algae, although pit connections link adjacent cells in most groups (*2*). In the red alga *Polysiphonia*, during development after fertilization, morphologically distinct pit connections, which I call "transfer connections," interconnect differentiating cell layers, and their role in enhancing cell-to-cell interactions would be consistent with developmental strategies observed in other multicellular organisms.

Red algal pit connections form between cells after incomplete cytokinesis, and their extracellular position and plug-like structure suggests that a role in intercellular transport is unlikely (*2, 3*). However, translocation of radioactively labeled compounds along files of cells has been reported (*4*), although there is no direct evidence that movement occurred through the pit connections. Structural differences in pit connections have been observed within different generations of a single organism (*3, 5*), and such modifications might effect intercellular transport, especially between cells of the reportedly parasitic carposporophyte.

After fertilization in *Polysiphonia*, a diploid generation (termed the carposporophyte) proliferates while attached to the female gametophyte (*6*). The carposporophyte consists of an outer, rapidly dividing layer of cells (the gonimoblast), of which some eventually develop into reproductive carpospores, and a central, irregularly shaped fusion cell which arises during early development and continues to expand outward by the gradual incorporation of adjacent gonimoblast cells. In addition, specific haploid cells of the female gametophyte, on which the diploid carposporophyte is borne, eventually establish cytoplasmic continuity with the fusion cell. It has frequently been suggested (*6, 7*) that the fusion cell itself, and those cells (both haploid and diploid) about to be incorporated into the fusion cell, provide nutritive material to the proliferating fringe of dividing goni-

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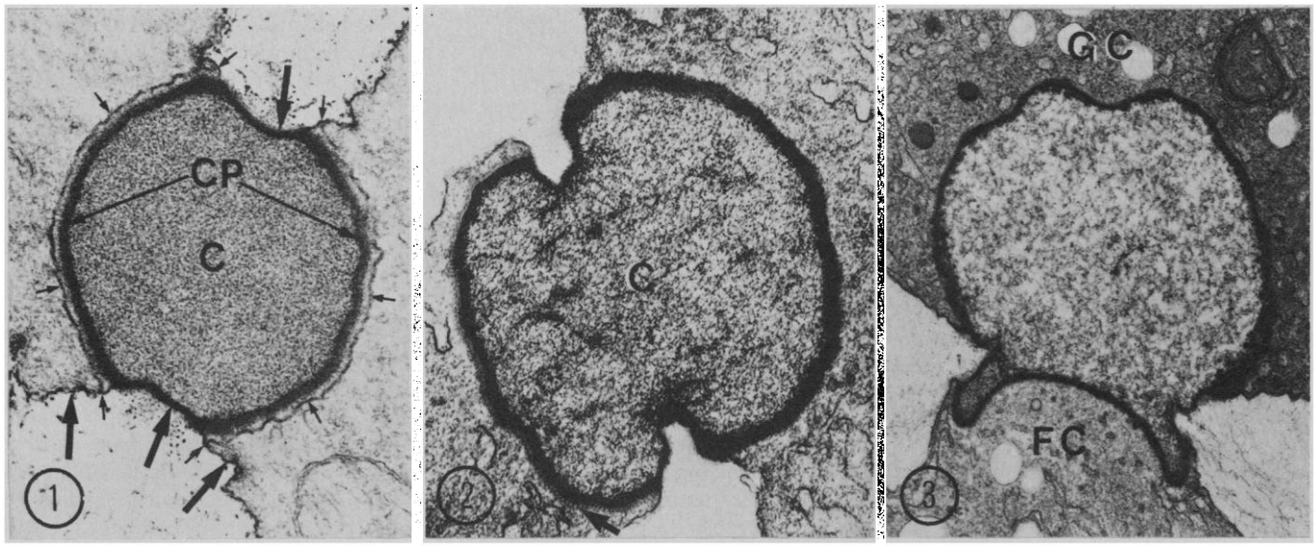

Fig. 1. Pit connection found between most cells of the thallus, consisting of a granular core (*c*) and electron dense caps (*cp*). The plasmalemma is continuous between cells (large arrows), and in addition, bifurcates to enclose the entire structure (small arrows) (× 48,420). Fig. 2. Transfer connection with a fibrous core (*c*) and cap composed of two layers. The outer layer is striated when viewed in the correct plane (arrow). The plasmalemma is continuous between cells but does not surround the connection as seen in Fig. 1 (× 50,500). Fig. 3. Low magnification micrograph of a transfer connection located between the fusion cell (*fc*) and a gonimoblast cell (*gc*) prior to fusion. Note the large size and flaring in one direction (× 18,200).

moblast cells and developing carpospores. In that these meristematic cells contain only proplastids with presumably little or no photosynthetic capacity, nutritive assistance would appear vital. The only course for translocation to the periphery would be through transfer connections which interconnect all cells of the carposporophyte plus those cells of the gametophyte which eventually fuse with it.

Plants of *Polysiphonia novae-angliae* Taylor were prepared for transmission electron microscopy in all stages of development (*8*). Pit connections occurring between most cells of *Polysiphonia* consist of a granular core and two electron opaque caps (Fig. 1). The plasma membrane is continuous between cells and, in addition, bifurcates to the exterior of each cap, enclosing the entire structure and isolating it from the cytoplasm (Fig. 1). Similarly structured pit connections have been observed in several other red algae (*2, 3, 5, 9*). An additional membrane possibly exists within, or associated with, the cap although freeze-fracture examination of pit connections of another red alga revealed only a single membrane in this position (*9*). In transfer connections between all cells of the carposporophyte, the plasma membrane is continuous between cells but is not observed to bifurcate and surround the structure (Figs. 2 and 3). Transfer connections therefore appear intracellular but, like normal pit connections, appear to restrict cytoplasmic continuity. Additional morphological differences consist of a less dense fibrous core, and caps composed of an inner darkly stained layer and an outer striated region that is only visible when sections are favorably oriented (Fig. 2). Although a membrane does not appear associated with these layers, the electron opacity of the cap might obscure such a structure.

During carposporophyte formation, transfer connections increase in size and flare out considerably into the cell most distant from the fusion cell. The largest transfer connections are found between the fusion cell and adjacent gonimoblast cells (Fig. 3). Since nutritive materials moving from the fusion cell toward the periphery would necessarily pass through these structures, an increase in the surface area of contact would clearly be advantageous. As translocation continued toward the periphery, flaring would become increasingly unnecessary in that fewer cells would be supplied through individual transfer connections. Transfer connections linking carpospores to the carposporophyte show only a slight increase in size, possibly because they are the end point of transport, and nutrients need not pass through them to supply additional cells.

The name transfer connection, chosen with a possible function in mind, refers only to those red algal pit connections whose morphology or position (or both) within a given thallus would suggest a role in enhancing intercellular transport. Cell-to-cell transport and communication is believed necessary for differentiation (*1*), and since many red algae have differentiated cell layers, transfer connections may be a common feature.

In many groups of red algae, cells of the carposporophyte have mature and apparently functional plastids throughout development; correspondingly, pit connections between these cells are similar to those separating all other cells of the thallus. *Polysiphonia*, however, is a highly advanced and differentiated red alga, and the coexistence of proplastids with transfer connections in certain cells of specialized regions of the plant may indicate the evolution of a post-fertilization scheme that requires nutritive transport toward the actively developing margins of the carposporophyte.

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20 November 1978; revised 22 February 1979