

## MODEL STUDIES ON THE INTERACTIONS AMONG CARBON SUBSTRATES, BACTERIA AND CONSUMERS IN A SALT MARSH ESTUARY

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### ABSTRACT

We have developed a lagrangian computer simulation model to investigate the interactions between dissolved and particulate carbon-substrates of detrital (vascular plants) and phytoplankton origin, microbes attached and free-living in the water column, and consumers of these substrates for a specific estuarine water body: the Duplin River, Sapelo Island, Georgia. Model simulation experiments were directed toward investigating the potential for both carbon resource and heterotrophic grazing as controls on free-living and particle-attached bacterial densities. Given available information from the literature and our own work, we present simulations of the model that approximate what one generally finds in this estuary during summer relative to the dynamics of particulate organic carbon (POC), dissolved organic carbon (DOC), free-living bacteria and microconsumers. Predicted dynamics of the bacteria were insensitive to macroconsumer grazing but highly sensitive to the parameter values governing microconsumer grazing rates and metabolism. Within reasonable constraints of the parameter values used, however, we were unable to reduce the predicted densities of particle-attached bacteria by manipulation of either resource (carbon) availability or grazing pressures. We propose several alternate hypotheses that might explain this discrepancy between model results and what is generally reported for natural systems.

In 1978 we reviewed the literature on the interactions between substrate, microbes and consumers of *Spartina* detritus in the water column of estuaries (Christian and Wetzel, 1978). Based on this we developed a five-compartment simulation model to examine within a theoretical framework those interactions seemingly most important. The results supported the conclusions of others at the time (Wiebe and Pomeroy, 1972) that an apparent dichotomy existed between the classical view of detrital food web dynamics (Darnell, 1967) and what was actually supported by contemporary information. Specifically, particles suspended in estuarine water are often, if not characteristically, devoid of microorganisms and yet microorganisms attached to particles are postulated (according to the paradigm) to be the major intermediary in estuarine detrital food web dynamics and a major carbon resource for macroheterotrophs. Alternate hypotheses which might explain the dichotomy are: (1) attachment of microorganisms to suspended particles is rare and microbes are not in fact major intermediaries in the food web, (2) heterotrophic grazing of microbes on particles and their turnover rates are so rapid that at any one time the standing stock of microbes is or can be very low and/or (3) free-living microbes play the role ascribed to attached forms.

Since our original report, a number of significant advances have been made which allow us to readdress this issue in a more quantitative fashion using a major revision of the original model. First, more realistic estimates of bacterial productivity have become available (Hagstrom et al., 1979; Karl, 1979; Fuhrman and Azam, 1980; Newell and Christian, 1981). Second, increased attention has been given to the role of microprotozoans in the cycles of essential elements and energy flow in aquatic ecosystems (Pomeroy, 1974; Sieburth and Davis, 1982; Robertson, 1983; Sherr and Sherr, 1983). Last, and specifically for our purposes here, research on the Duplin River estuary adjacent to Sapelo Island, Georgia, has progressed to allow for more realistic simulation modeling studies of detrital

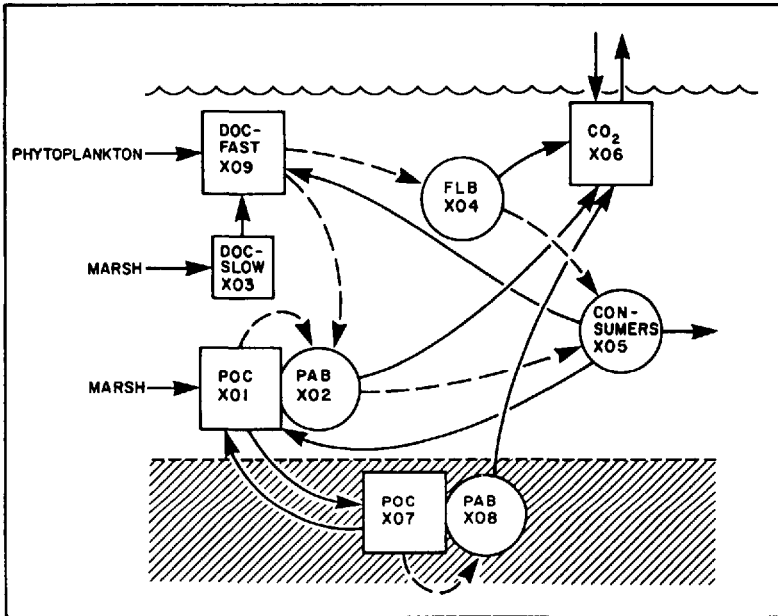


Figure 1. The conceptual diagram of compartment and flow structure used to simulate the interactions and dynamics of carbon in the detrital based food web of the Duplin River estuary, Sapelo Island, Georgia. See text for a detailed description of the model compartments and mathematics.

food web interactions (Newell and Christian, 1981; Pomeroy and Wiegert, 1981; Imberger et al., 1983; Sherr and Sherr, 1983).

In this paper, we present a major revision of the original model, compare it to new information relative to the ecology of the Duplin River estuary, and report the results of model simulation studies designed to address two potential controls on detrital-carbon dynamics and microbial energetics. First, we examine the influence of organic carbon sources (both particulate and dissolved) on particle-attached and free-living bacteria growth. Second, we examine the effects of heterotrophic grazing by net zooplankton, microprotozoa and microprotozoa experiencing predation as potential controls on bacterial density.

### SIMULATION MODEL

*Compartmental and Mathematical Structure.*—We expanded on the original model by (1) increasing structural (both compartmental and mathematical) complexity, (2) time-scaling the model to field and experimental measurements for a specific system (the Duplin River, Sapelo Island, Georgia) and (3) incorporating real concentration and rate data reported in the literature.

Figure 1 shows the compartmental and flow structure of the simulation model. Dashed lines indicate a flux that is described by a non-linear, feedback controlled equation. A solid line indicates a flux that is described by a linear, donor controlled equation. The mathematical structure of the model follows the approach originally described by Wiegert (1973; 1975). The functional forms of the equations and a detailed explanation of their formulation can be found in Christian and Wetzel (1978).

Linear functions are used to simulate exports from the system and metabolic processes such as respiration, egestion and excretion. The non-linear, feedback-controlled functions are used to describe the flux of carbon between abiotic → biotic or biotic → biotic pathways where either the donor or

recipient compartment can limit exchange. As for our previous model, we modified the general form of the resource control feedback expressions for particle-attached bacteria to represent potential limitation due to either surface area available for attachment and/or biochemical availability. For clarity, it is important to point out relative to the mathematical structure of the feedback terms that the expressions are functions of both threshold and limiting concentrations. That is, for donor or resource feedback control, there is a threshold concentration below which the resource becomes limiting and a lower concentration at which it becomes unavailable for uptake. Likewise for recipient or self-controlled feedback, there is a threshold concentration (density or biomass) above which space or some space-related factor limits the rate of growth and an upper limit that defines the maximum maintainable concentration. Although, the model has been designed to simulate characteristics and processes occurring in the Duplin River estuary, the conceptual and mathematical structures are general. For this model, concentrations are mg C liter<sup>-1</sup> and flows are mg C liter<sup>-1</sup> h<sup>-1</sup>.

For computer analysis, the model was programmed in FORTRAN 77 and run on a PRIME 850 mainframe. The Euler method with an iteration interval of 0.1 h (Wiegert and Wetzel, 1974; Wetzel and Wiegert, 1983) was used for digital solution of the equations written in differential form. A copy of the program is available on request from one of us (R.L.W.).

## MODEL DATA AND COMPARTMENT DESCRIPTION

*Site Description and Hydrology.*—The Duplin River is a 12.5 km long tidal lagoon west of Sapelo Island, Georgia. The watershed is approximately 11 km<sup>2</sup> and composed almost entirely of intertidal *Spartina alterniflora* Loisel salt marshes. Freshwater input to the river is minimal and restricted to rainfall. Considerable research has been conducted in this watershed since the early 1950's and the results have been recently summarized by Pomeroy and Wiegert (1981).

The hydrology of the Duplin River has been described by Ragotzkie and Bryson (1955) and more recently by Imberger et al. (1983). There are three tidal prisms within the river and the estimated mixing time during neap tides for the upper prism is 89 h. For the model simulation studies, we conducted each model run for 144 h by extrapolating this mixing time estimate to include the entire river reach. Thus, water entering the head of the Duplin from the marshes has an approximate residence time of 144 h and time in the model is comparable to tracking downriver water movement. The implicit (and unconfirmed) assumption in the model is that suspended particulates (POC, bacteria and consumers) are transported as are dissolved constituents. All simulations of the model began at midnight on a neap high tide during the summer at the headwaters of the Duplin River.

*Data Input and Parameter Value Justification.*—In the following we describe the source for each of the model parameter values. The values given are for the nominal simulations. Later we describe how these were modified for the simulation experiments. Although this undoubtedly adds length and certainly some tedium to reading, we consider it necessary for a full understanding of the approach and final conclusions we offer.

**PARTICULATE ORGANIC CARBON (POC: X01).** Particulate organic carbon concentrations in the Duplin River were reported by Imberger et al. (1983) and have been measured by one of us (R.R.C.). In both studies, POC concentrations in the headwaters were approximately 20 mg C liter<sup>-1</sup>. Concentrations decreased to approximately one-half this at the mouth (Imberger et al., 1983). While POC is a combination of both living and nonliving material, we assumed for model initial conditions that POC available for microbial attachment and utilization is 20 mg C liter<sup>-1</sup>. Thus initially, the POC entering the headwaters is considered nonliving. POC concentration as well as microbial activity in the water column also appears dependent on tidal stage (Hanson and Wiebe, 1977; Imberger et al., 1983). POC is deposited during slack water and resuspended during peak ebb and flood periods. We assumed that the apparent tidal signal in Hanson and Wiebe (1977) glucose uptake data results from the deposition and resuspension of particles with attached bacteria. This cycle was simulated using a cosine function with a second order decay for exchange between water column and sediments to allow permanent deposition of a small fraction of the POC. Because of the semi-diurnal tidal regime of the Duplin, the suspension-deposition period was 3 h.

The availability of POC to microbes involves both surface area for attachment and metabolic (biochemical) availability as an energy source. We estimated the surface area per mg POC by microscopy of Duplin River water samples. The average surface area was  $1.4 \times 10^{10}$   $\mu\text{m}^2$  mg C<sup>-1</sup> although complex internal surfaces were not measured. Relative to the time scale for model simulation, we assumed that no "aging" of detritus occurred and that only a small fraction of the POC represented organic material that was metabolically available to the microbes. However, we allowed most to be degraded at a limited rate. The labile POC fraction was set at 0.001 g C g C-POC<sup>-1</sup> or 0.1% of the initial POC concentration. This agrees with estimates of availability from studies on the summer rates of *Spartina alterniflora* decomposition using litter bags (Christian, 1984). Truly refractory (i.e., un-

available) POC was set at 20% of the initial POC concentration based on lignin content of *S. alterniflora* detritus (Hodson et al., manuscript submitted).<sup>1</sup>

**PARTICLE ATTACHED BACTERIA (PAB: X02).** Newell and Christian (1981) described the frequency of attached bacteria relative to free-living bacteria in samples from near the head of the Duplin River. Also, Christian (unpubl. data) estimated the standing stock of PAB at 0.066 mg bacterial-C liter<sup>-1</sup> or 0.003 mg bacterial-C mg POC<sup>-1</sup>. This represents approximately one bacterium per 68  $\mu\text{m}^2$  of particle surface area. We assumed that bacteria would be maximally attached (i.e., reached maximum maintainable density per particle and limited by space for further growth) when one cell is found per  $\mu\text{m}^2$  or 0.16 mg bacterial-C mg POC<sup>-1</sup>. The spatial threshold response density at which limitation begins was set at one-half this maximum or 0.08 mg bacterial-C mg POC<sup>-1</sup>. These values are much higher than what is seen in nature and are considered as potentials only. The possibility remains that limitations may arise at lower values as a result of biological interactions separate from these physical constraints.

The maximum growth rate of attached bacteria was not measured directly. In the absence of data on particle-attached bacteria growth, maximum potential growth of PAB was assumed to equal that of free-living cells. The maximum growth rate for PAB was set at 0.9 h<sup>-1</sup> the origins of which will be discussed later. Aerobic growth efficiency of bacteria may vary as a function of realized growth but it has most consistently been measured as approximately 0.6 g C g C<sup>-1</sup> (Payne, 1970; Christian and Wetzel, 1978). Based on this, microbial respiration was set at 0.4 gC gC<sup>-1</sup> of assimilated carbon and the maximum specific assimilation rate was set at 1.2 g C g C<sup>-1</sup> h<sup>-1</sup>. These parameter values were used for bacterial utilization of both POC and DOC-F (X09). Rates measured in the field are considerably less than this (Newell and Christian, 1981). We retained these higher values in the model for simulation because the negative feedback structure of the equations should predict the lower realized growth if the controls are accurately represented; precisely the purpose of the model simulation studies. For all nominal simulation model runs, the preference for each substrate was assumed equal when either was not limiting (i.e., the structure of the equations allow for the recipient to switch resources depending on an assigned preference value and its availability).

**DISSOLVED ORGANIC CARBON (DOC-S; X03 AND DOC-F; X09).** Concentrations of DOC along the axis of the Duplin River were reported by Imberger et al. (1983). Highest concentrations were in the headwaters and decreased exponentially downriver. The authors proposed that the majority of the DOC measured was refractory but an unknown fraction was rapidly recycled. Earlier work by Sottile (1973) supports this conclusion. We have assumed that the major source of DOC to bacteria is rapidly recycled forms and not represented by the bulk measurements. To represent this in the model, we partitioned DOC into two classes; a slow (DOC-S) and a fast (DOC-F) pool, driven by different inputs.

Two sources of "fast" DOC are considered. The first source was from phytoplankton release and the second was from the conversion of a "slow" DOC pool to the labile, DOC-F fraction. Summer phytoplankton primary productivity in the Duplin River was assumed to be 0.6 mg C liter<sup>-1</sup> integrated over a 14-h photoperiod (Pomeroy et al., 1981). We converted this estimate to a specific hourly rate by assuming the rate followed a sine wave function over the photoperiod. Phytoplankton DOC release was then calculated as a fraction of the estimated hourly rate. Thomas (1971) concluded that no more than 7% of the primary production was lost as DOC in Georgia estuaries although higher values have been reported (Azam et al., 1983). For nominal simulation runs, the 7% value was used.

For the second source, the conversion of slow to fast DOC was estimated from Sottile (1973). In a 47-day microcosm experiment, Sottile found a loss of 1.73 mg DOC liter<sup>-1</sup>. This was assumed to be the result of a slow to fast conversion (hydrolysis) and subsequent mineralization. This equals a rate of 0.0015 mg C liter<sup>-1</sup> h<sup>-1</sup> (assuming exponential decay) which we set as a constant input for the duration of each simulation run.

**FREE-LIVING BACTERIA (FLB: X04).** The initial conditions for FLB were derived from acridine orange direct counts of samples taken at the headwaters of the Duplin (Newell and Christian, 1981). Growth parameters were derived from the study of Christian et al. (1982). In their study, the growth of a mixed, natural assemblage of bacteria under different nutrient conditions was compared. The specific growth rate at each nutrient concentration was plotted against nutrient concentration added (25 to 100 mg yeast extract liter<sup>-1</sup>) to a 3  $\mu\text{m}$  filtrate of estuarine water (water temperature for the experiments was 29°C). Assuming Michaelis-Menten kinetics, the estimated maximum specific growth rate ( $\mu_{\text{max}}$ ) was 0.9 h<sup>-1</sup> and the maximum bacterial density was  $1 \times 10^{11}$  cells liter<sup>-1</sup>. Based on these data, we set the maximum possible free-living bacterial biomass at 3.5 mg C liter<sup>-1</sup> and assumed that limitation to growth started at 50% of this value, i.e., 1.8 mg C liter<sup>-1</sup>. Again, these values provide the upper bounds for bacteria growth under optimum conditions and not what would be expected from field measures.

<sup>1</sup> Hodson, R. E., R. R. Christian and A. E. Maccubbin. Lignocellulose and lignin in the salt marsh grass, *Spartina alterniflora*: initial concentration and short term post-depositional changes in detrital material. Mar. Biol. (submitted).

The feedback control parameters for resource limitation were also derived from this study. The carbon content of yeast extract was assumed 50% by weight and twice the derived  $k_m$  was 40 mg DOC-F liter<sup>-1</sup>. Thus, the DOC-F concentration below which limitation begins was set at 40 mg C liter<sup>-1</sup>. We further assumed that a DOC-F concentration that would theoretically support a specific growth rate of 0.001 h<sup>-1</sup> (approximately 1 month generation time) would be insufficient to maintain viability of the cells. This DOC-F concentration based on a modified Lineweaver-Burke transformation of the data was 0.02 mg DOC-F liter<sup>-1</sup>. The feedback function was calculated as a power function to simulate more closely what appears to occur naturally.

**CONSUMERS (CONS: X05).** The model simulation experiments investigated two types of consumer populations. For the first, we assumed the consumer was a macroconsumer and energetically typified by net zooplankton. To our knowledge there is little data for these populations in the Duplin River and we used parameter values for growth, metabolism and the feedback functions directly from Kremer and Nixon (1978). We included these macroconsumer model studies to test the classical view of detrital food web dynamics in light of the revised model structure and new data.

For the second series of experiments, we assumed the consumer population was represented by a microconsumer and best modeled after microprotozoa. There is a fundamental difference relative to ingestion and assimilation of substrates between micro- and macroconsumer groups. For macroconsumers, whole particles are ingested or water filtered and thus ingestion and assimilation of either PAB or FLB is a passive process. For microconsumers ( $\mu$ -flagellates in this case), ingestion is an active process analogous or more comparable to classical predator-prey interactions. For each series of experiments, we derived the model ingestion equations to represent these two fundamentally different strategies. Also, for the last series of microconsumer simulation experiments, we investigated the effect of imposing a second-level consumer on  $\mu$ -flagellates by artificially removing a fraction of the microconsumer population each iteration interval.

The initial conditions for  $\mu$ -flagellate biomass were derived from Sherr and Sherr (1983). Biomass was estimated as  $4.5 \times 10^{-3}$  mg C liter<sup>-1</sup>. The maximum growth rate for the population was set at 0.125 h<sup>-1</sup> based on work with *Monas* sp. reported by Sherr et al. (1983). From their studies, Sherr et al. (1983) estimated a net growth efficiency of 30% which corresponds to a maximum specific ingestion rate of 0.4 h<sup>-1</sup> and a respiration rate of 0.7 times the ingestion rate. We assumed for the nominal model runs that  $\mu$ -flagellates could realize the maximum specific growth rate from either PAB or FLB (i.e., preference for these substrates were equal for non-limiting resource conditions).

Density dependent limitations on  $\mu$ -flagellate growth were taken from the work of King et al. (1980), Fenichel (1982a; b), Rassoulzadegan (1982) and Azam et al. (1983). Maximum densities of  $\mu$ -flagellates appear to range from  $1 \times 10^6$  to  $6 \times 10^6$  cells liter<sup>-1</sup>. From these data and estimates of cell size and carbon content, we estimated the maximum maintainable standing stock as 0.45 mg C liter<sup>-1</sup>. Based on Fenichel (1982b), the threshold response density was set at 0.25 mg C liter<sup>-1</sup>.

The parameters governing resource control feedback limitation were estimated from Sherr et al. (1983). The threshold concentrations for both feedback functions were set at 10 mg C liter<sup>-1</sup>. The refuge level or bacterial biomass below which they are not available for ingestion was assumed to be less than that reported by both Sherr et al. (1983) and Fenichel (1982a). Their feeding experiments were conducted with bacterial densities greater than  $10^9$  cells liter<sup>-1</sup>. We assumed a refuge level for free-living bacteria of  $5 \times 10^8$  cells liter<sup>-1</sup> which corresponds to 0.017 mg C liter<sup>-1</sup>. For PAB, we assumed that their availability for microconsumer ingestion was a function of their density on particles and the resource control feedback was estimated as one minus the PAB spatial control feedback term. Also, the self-control feedback term for microconsumers was calculated as a power function (as for bacteria) rather than a linear function of density.

**OTHER COMPARTMENTS.** The compartments representing POC and PAB in the sediments and CO<sub>2</sub> in the water were tracked for mass balance calculations only. While PAB resided in the sediments carbon metabolism (respiration) continued, but the compartment was removed from any grazing pressure. Because bacterial numbers are generally two or more orders of magnitude greater in sediments than the water column, we feel this assumption does not compromise the predicted dynamics while residing in the water column (i.e., suspended). The carbon dioxide compartment was tracked to allow calculation of total metabolism for the period of simulation.

**Model Simulation Experiments.**—The model simulation experiments are summarized in Table 1. Four sets of computer runs were made to simulate the effects of (1) carbon resource allocation by particle-attached bacteria, (2) sensitivity of the microbial compartments to various DOC inputs, (3) potential controls via grazing by an assumed macroheterotrophic consumer and (4) the potential controls on carbon dynamics by microheterotrophic consumers. We based our analyses of these model simulation experiments on the change in compartmental dynamics and/or flows relative to nominal predictions (i.e., compartmental dynamics and/or flows predicted by simulations using the input data discussed previously). Parameter values were changed one at a time with all others set to their nominal value except where they were known to covary.

Table 1. A summary of all model simulation experiments to test compartment responses to variation in microbial resource partitioning, DOC input, macroconsumer grazing and microconsumer energetics

Experiment	Model parameters	Parameter values
POC vs. DOC		
POC preference	P0102	1.0, 0.75, 0.25, 0.0
DOC preference	P0902	0.0, 0.25, 0.75, 1.0
DOC input		
Phytoplankton	F0009	3.5, 14, 25, 50%
Marsh	F0003	0.0, 2×, only
Macroconsumer grazing		
POC-PAB preference	P0105	1.0 (only source)
POC-PAB ingestion	T0105	2× and 0.5× nominal
Microconsumer energetics		
PAB preference	P0205	1.0, 0.75, 0.25, 0.0
FLB preference	P0405	0.0, 0.25, 0.75, 1.0
Ingestion	T0205, T0405	2× and 0.5× nominal
Assimilation efficiency	AE0205, AE0405	0.75, 0.50
FLB resource limitation	A0405	10 <sup>6</sup> , 10 <sup>7</sup> cells

## RESULTS AND DISCUSSION

*Simulations with Macroconsumers.*—The first series of simulation experiments were done assuming that grazing of bacteria and POC was by zooplankton. The results of the nominal run are shown in Figure 2. This run embodied the best estimates for each of the parameter values obtained from the literature and our own work. The strong tidal signal in POC and PAB dynamics is apparent. Both particle-attached and free-living bacteria increased in biomass and hence density with macroconsumer grazing at rates reported by Kremer and Nixon (1978). Particle-attached bacteria grew rapidly to the maximum density set by the self-control feedback equation which corresponds to a cell density of approximately one cell per  $\mu\text{m}^2$ . Based on direct counts from the Duplin River, this is 50 to 100 times greater than observed on naturally occurring particles. Free-living cells increased linearly with damped oscillations due to the discontinuous input of DOC from phytoplankton. The interesting result relative to macroconsumer dynamics is that under what we assume nominal conditions, the population exerts no control over bacterial growth or steady state biomass and perhaps more importantly, cannot survive (i.e., meet energy maintenance costs) or grow on detrital, particle-attached or free-living microbial carbon sources alone.

By doubling the ingestion rate of macroconsumers for all resources, macroconsumers survived and grew but the actual rate of ingestion needed is unrealistically high relative to published data. Even with this ingestion rate and consumer growth, both FLB and PAB dynamics were similar to the nominal case. Halving the ingestion rate (i.e., to a rate more typically reported) resulted in the predicted population standing stock being reduced to essentially zero. Figure 3 illustrates these simulation experiments and the results suggest that the microbial compartments are insensitive in large degree to macroconsumer grazing and that macroheterotrophs require carbon sources other than or certainly in addition to detrital or bacterial origins.

These results might be expected if bacteria are not resource (carbon) limited. The possibility that they are substrate limited was examined in several ways. First,

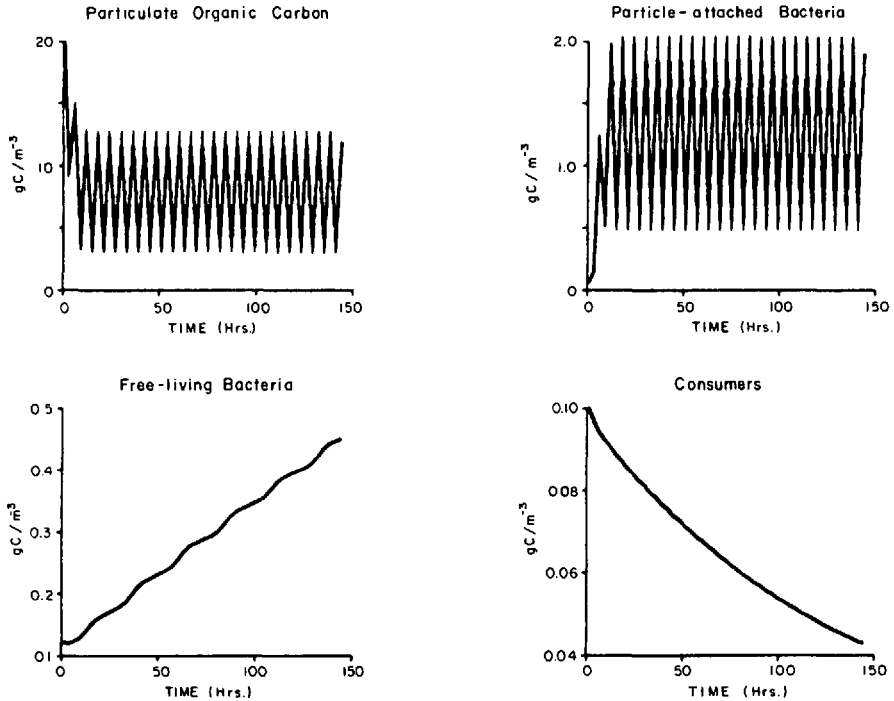


Figure 2. Simulated dynamics of carbon for the four principal compartments of the model with nominal macroconsumer grazing pressure.

the dynamics of the DOC compartment appeared reasonable (based on Duplin River data). Slow-DOC decreased by only 3% over the 144 h simulations. Fast-DOC concentrations were cyclical due to the input functions, and predicted concentrations were below detection limits by conventional means. These results are consistent with the results of Sottile (1973). Secondly, computed generation times and bacterial productivities were very similar to the results of Newell and Christian (1981) for the Duplin River in summer. Model predictions for the bacterial compartments thus appear consistent with what one would expect under low grazing pressures.

The rapid growth response and predicted density of PAB might still be the result of inadequacies in our representation of PAB-carbon resource interactions even though behavior with respect to DOC seems realistic. We assumed in these model simulations that the metabolism of PAB and FLB are the same. If this is a realistic assumption, PAB should respond to variable DOC inputs in a manner similar to FLB. Thus, if a problem exists in the dynamics of PAB, it may reside in either (1) inadequate or unrealistic depiction of POC as a PAB carbon resource or (2) differences in metabolism between PAB and free-living cells.

To address the first possibility, we varied the preference of PAB for either POC or DOC. The extremes are represented by either resource being the sole carbon input to PAB. The results were that there was no significant change in either PAB growth or density even when DOC was the sole carbon source. Thus, the rapid growth and what would appear unrealistic, bacterial densities on particles could not be ascribed solely to POC or DOC as resources for PAB.

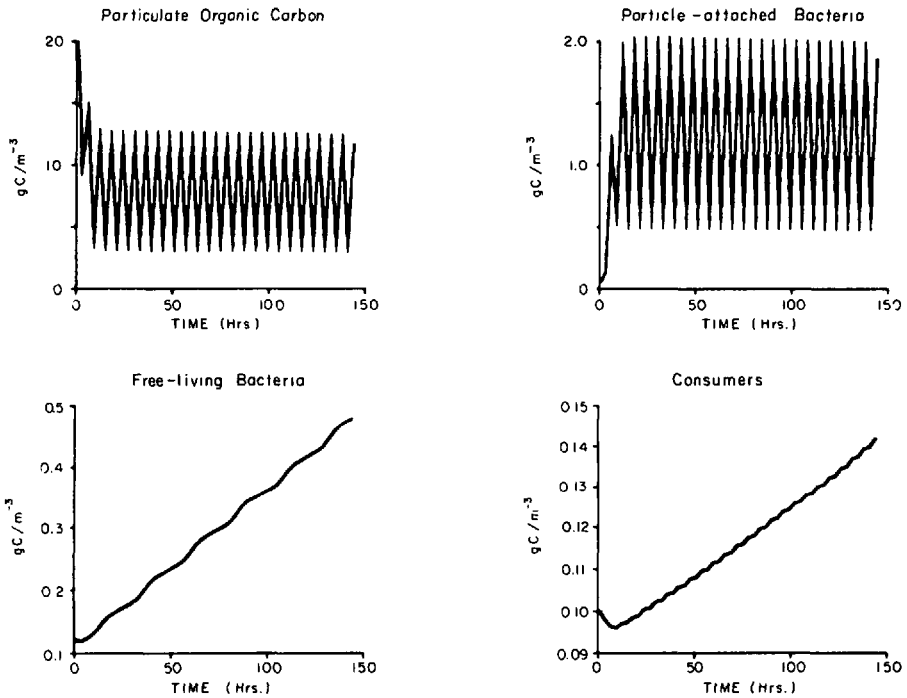


Figure 3. Simulated dynamics of carbon for the four principal compartments of the model with the macroconsumer ingestion rate set at twice the nominal value.

Relative to the second possibility, D. Rice and R. B. Hanson and R. C. Newell (this issue) suggest that the metabolism of PAB may differ from FLB. Newell found that growth efficiencies of bacteria were dependent on substrate quality. Thus, PAB may have lower growth efficiencies than FLB. Rice and Hanson suggest that PAB may produce considerable amounts of extracellular products in the form of slime or extracellular proteins. These differences in metabolic characteristics were not investigated via model simulation and require further study.

*Simulations with Microconsumers.*—The results of the nominal simulation with microconsumers are shown in Figure 4. For these model studies, grazers are primarily  $\mu$ -flagellates as recently described for the Duplin River by Sherr and Sherr (1983). POC dynamics are similar to the previous results however bacterial dynamics are markedly different. PAB initially grew to the maximum standing stock (density on particles) then declined to 40% of the maximum. This lower density however still corresponds to bacterial densities on particles greater than commonly observed in the Duplin River but reported in other literature. FLB were reduced to a density approximately twice the set refuge level. In order to achieve these controls on bacterial densities, the microconsumer compartment grew to inordinately high standing stocks as a result of no other control on their growth (i.e., growth of the compartment for these simulations could only be controlled by resource and/or spatial limitation). Thus, for these simulations the bacteria were sensitive to the grazing pressure of a microconsumer population unchecked by other controls. The results suggest that under the given conditions,

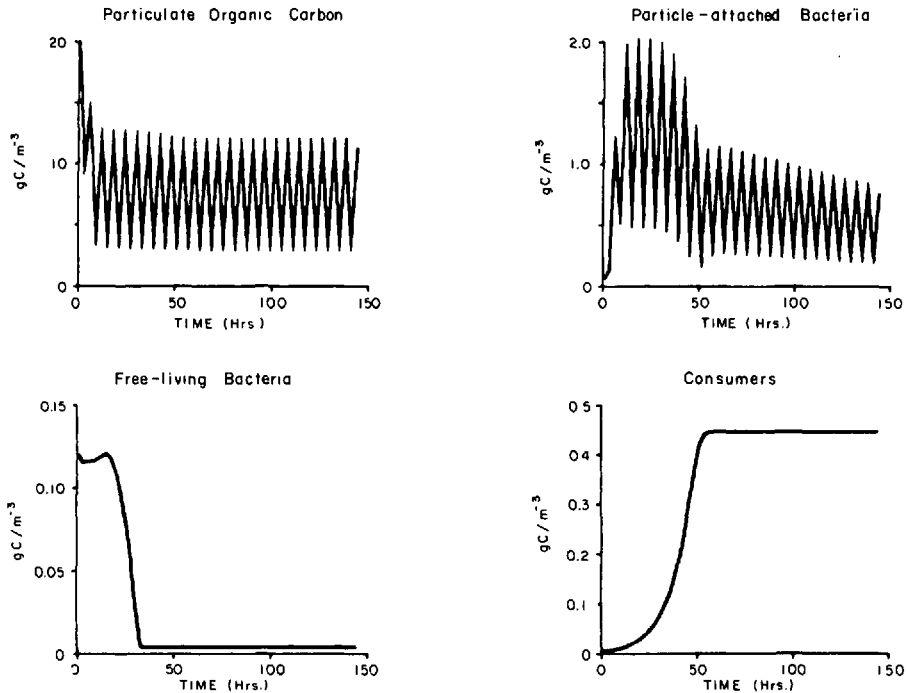


Figure 4. Simulated dynamics of carbon for the four principal compartments of the model with nominal microconsumer grazing pressure.

$\mu$ -flagellate grazing (using literature values) can control bacterial density and as importantly bacteria alone can support their growth.

The potential control of microbial dynamics by microconsumers were further evaluated in several ways. Simulation experiments were conducted in the absence of predation on microconsumers in which ingestion rates, assimilation efficiencies and resource preferences of the microconsumers were varied. Figure 5 shows the results of doubling the ingestion rate and reducing the assimilation efficiency from the nominal value of 95 to 75%. This is the only simulation experiment out of 45 that effectively reduced the densities of PAB. As for the nominal case, the microconsumer compartment grew to unrealistic densities (i.e., relative to field observations). In contrast to macroconsumer grazing, the bacterial compartments were very sensitive to relatively minor changes to the parameter values governing microconsumer growth and metabolism (Table 2). Our understanding of microprotozoan bioenergetics is just developing. Based on these model studies, it is evident that a much better understanding of their physiological ecology and population dynamics is necessary to better address their role in detrital-based food webs.

*Simulations of Resource Partitioning by Bacteria.*—Numerous simulations were run with either macroconsumer or microconsumer grazing in which resource availability to the bacteria was manipulated. Resource preference by PAB, phytoplankton DOC release and hydrolysis of slow-DOC to fast-DOC were all evaluated relative to overall compartmental behavior. Resource preference by PAB

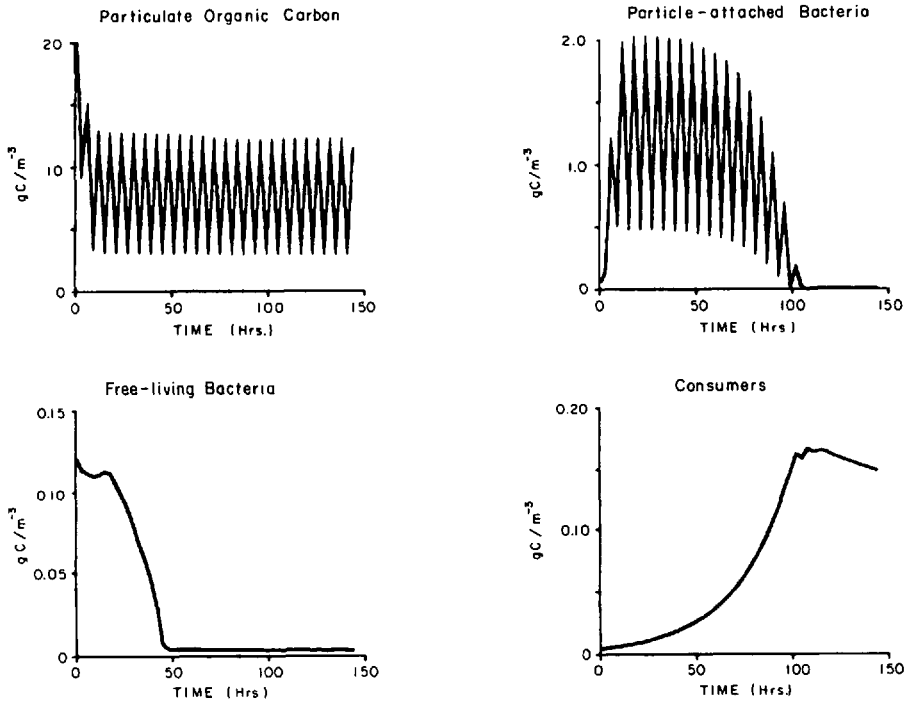


Figure 5. Simulated dynamics of carbon for the four principal compartments of the model with the microconsumer ingestion rate set at twice the nominal value and the assimilation efficiency set at 75%.

had little effect on compartmental dynamics. Increasing phytoplankton DOC release from 7 to 14%, 25 or 50% of photosynthesis or doubling the hydrolysis rate produced effects dependent on the grazing pressure. For macroconsumer experiments, increased DOC availability had no effect on PAB but increased the growth of FLB. For microconsumer experiments, increased DOC availability increased PAB but failed to overcome the grazing pressures on FLB relative to increasing FLB standing stocks. Overall, it would appear that the control on bacteria by grazing exceeds that due to carbon resource availability within this model context.

*Simulations with Predation on Microconsumers.*—Obviously, predation on the consumers described in the model operates as a potential control. Given the lack of influence that macroconsumers apparently exercise in this model, it is unlikely that predation on them directly influences microbial dynamics. However, the same cannot be concluded relative to predation on microconsumer populations. The principal line of evidence suggested by the simulation experiments is that given what appear reasonable estimates for the parameter values governing microconsumer growth,  $\mu$ -flagellate standing stocks or densities predicted by the model are rarely observed in nature. To investigate the potential role of a second level predator on microconsumers, we artificially imposed a predator drain on the compartment by removing a fixed percentage of the biomass per iteration.

The final standing stocks (i.e., at 144 h) of PAB, FLB and microconsumers are

Table 2. Model simulation experiments on the effects of changing ingestion rate, assimilation efficiency and resource preferences of microconsumers on the predicted microbial compartment densities

Ingestion rate	Assimilation efficiency (%)	Resource preference		PAB	FLB
		PAB	FLB	(mg C liter <sup>-1</sup> )	
0.40	95	50	50	0.760	0.0037
0.40	75	50	50	1.820	0.0039
0.40	50	50	50	1.920	0.4200
0.80	75	50	50	0.003	0.0033
0.20	75	50	50	1.900	0.0340
0.40	75	100	0	1.660	0.4300
0.40	75	75	25	1.760	0.2000
0.40	75	25	75	1.850	0.0038
0.40	75	0	100	1.920	0.0046

given in Table 3 for these simulation experiments. Increased predation on microconsumers slightly increased PAB density. FLB were released from their refuge limit control by low microconsumer predation (1% per day) and changed slowly with predation pressures to 25% per day. A predation rate of 25% per day maintained microconsumers at near natural densities and FLB at approximately 3 × natural densities. Whereas doubling the ingestion rates of microconsumers and imposing a 10% per day predatory loss maintained FLB at near natural densities but allowed for a 12 × increase in microconsumer standing stocks. Again, we infer from these results that more precise knowledge of microconsumer dynamics both spatially and temporally is essential to understanding these interactions and potential controls.

### CONCLUSIONS

Using available data and information from the literature and our own work, we have produced model simulations that approximate what one finds in the Duplin River estuary during summer with respect to the reported dynamics of POC, DOC, free-living bacteria and microconsumers. Within reasonable constraints of parameter values, however, we have been unable to reduce the predicted standing stocks (densities) of bacteria attached to POC to observed concentrations by either altering carbon resource availability or grazing pressures.

There are several alternative explanations for the apparent dichotomy between model predictions and what is observed in nature relative to the numbers of

Table 3. Model simulation experiments on the effects of adding a secondary level consumer (i.e., predation) on the microconsumer compartment

Ingestion rate (%)	Assimilation efficiency (%)	Predation rate (% per day)	PAB	FLB	Consumer
			(mg C liter <sup>-1</sup> at 144 h)		
0.40	75	0.0	1.82	0.004	0.038
0.40	75	1.0	1.86	0.250	0.023
0.40	75	5.0	1.87	0.270	0.018
0.40	75	10.0	1.87	0.290	0.014
0.40	75	25.0	1.89	0.340	0.006
0.80	75	10.0	1.53	0.140	0.053

bacteria on particles. First, the metabolism of particle-attached bacteria may be sufficiently different from free-living cells as to deter rapid colonization of particles. Factors for this might include decreased growth yields of bacteria utilizing detrital-POC substrates (see Newell, this issue) and production of extracellular material by bacteria which acts as an energy expenditure not resulting directly in cell growth (see Rice and Hanson, this number). Although growth rate estimates of attached bacteria by the frequency of dividing cells (FDC) technique have been used to infer rapid growth of PAB (Newell and Christian, 1981), recent findings for FDC's of bacteria in sediments suggests that attached bacteria may have slower growth rates than predicted by FDC (Fallon et al., 1983). Second, we have intentionally eliminated exchange between PAB and FLB in the model. This was done to more directly evaluate the dynamics of each. Jacobsen and Azam (this issue) have demonstrated that bacteria can be readily lost from particles presumably as daughter cells. The model does not account for this as all PAB growth remains with the particle. If a high proportion of daughter cells of PAB become FLB, the predicted PAB growth would be much less.

One last possible explanation for the apparent dichotomy may be (with tongue in cheek) that the model predictions are in general correct and that our field and laboratory techniques have been insufficiently designed. Although we consider this an unlikely explanation, consider the following. Bacteria are known to attach both reversibly and irreversibly to surfaces (Marshall, 1976). The techniques most often used to enumerate bacteria attached to particles are not conducive to the retention of reversibly sorbed cells: i.e., fixation in preservative, shaking and filtration. If attachment is largely a reversible reaction at the cell surface, the possibility exists that attachment as presently judged from field samples may be underestimated.

In a more general sense, the model we have presented and discussed was developed with particular regard for spatial and temporal scales of measurement. We have attempted in these simulations to match both scales with the biological phenomenon (rate processes and turnover times) investigated through simulation studies. Based on this experience, we suggest that failure to have done so would have severely compromised the usefulness of the exercise.

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#### LITERATURE CITED

- Azam, F., T. Fenchel, J. G. Field, J. S. Gray, L. A. Meyer-Reil and F. Thingstad. 1983. The ecological role of water-column microbes in the sea. *Mar. Ecol. Prog. Ser.* 10: 257-263.
- Christian, R. R. 1984. A life table approach to decomposition studies. *Ecology* 65: 1693-1697.
- and R. L. Wetzel. 1978. Interactions between substrate, microbes, and consumers of *Spartina*

- detritus in estuaries. Pages 93–113 in M. Wiley, ed. *Estuarine interactions*. Academic Press, New York.
- , R. B. Hanson and S. Y. Newell. 1982. Comparison of methods for measurement of bacterial growth rates in mixed batch cultures. *Appl. Environ. Microbiol.* 43: 1160–1165.
- Darnell, R. M. 1967. Organic detritus in relation to the estuarine ecosystem. Pages 376–382 in G. H. Lauff, ed. *Estuaries*. Amer. Assoc. Adv. Sci., Washington, D.C.
- Ducklow, H. W. 1982. Chesapeake Bay nutrient and plankton dynamics. 1. Bacterial biomass and production during spring tidal destratification in the York River, Virginia, estuary. *Limnol. Oceanogr.* 27: 651–659.
- Fallon, R. C., S. Y. Newell and C. S. Hopkinson. 1983. Bacterial production in marine sediments: will cell-specific measures agree with whole-system metabolism. *Mar. Ecol. Prog. Ser.* 11: 119–127.
- Fenchel, T. 1982a. Ecology of heterotrophic microflagellates. II. Bioenergetics and growth. *Mar. Ecol. Prog. Ser.* 8: 225–231.
- . 1982b. Ecology of heterotrophic microflagellates. IV. Quantitative occurrence and importance as bacterial consumers. *Mar. Ecol. Prog. Ser.* 9: 35–42.
- Fuhrman, J. A. and F. Azam. 1980. Bacterioplankton secondary production estimates for coastal waters of British Columbia, Antarctica and California. *Appl. Environ. Microbiol.* 39: 1085–1095.
- Hagstrom, A., U. Larsson, P. Horstedt and S. Normark. 1979. Frequency of dividing cells, a new approach to the determination of bacterial growth rates in aquatic environments. *Appl. Environ. Microbiol.* 37: 805–812.
- Hanson, R. B. and W. J. Wiebe. 1977. Heterotrophic activity associated with particulate size fractions in a *Spartina alterniflora* Loisel. salt marsh-estuary, Sapelo Island, Georgia, and the continental shelf waters. *Mar. Biol.* 42: 321–330.
- Imberger, J., T. Berman, R. R. Christian, E. B. Sherr, D. E. Whitney, L. R. Pomeroy, R. G. Wiegert and W. J. Wiebe. 1983. The influence of water motion on the distribution and transport of materials in a salt marsh estuary. *Limnol. Oceanogr.* 28: 201–214.
- Karl, D. M. 1979. Measurement of microbial activity and growth in the ocean by rates of stable ribonucleic acid synthesis. *Appl. Environ. Microbiol.* 38: 850–860.
- King, K. R., J. T. Hollibaugh and F. Azam. 1980. Predator-prey interactions between the larvacean *Oikopleura dioica* and bacterioplankton in enclosed water columns. *Mar. Biol.* 56: 49–57.
- Kremer, J. N. and S. W. Nixon. 1978. A coastal marine ecosystem. Springer-Verlag, New York. 217 pp.
- Marshall, K. C. 1976. Interfaces in microbial ecology. Harvard University Press, Cambridge, Mass. 156 pp.
- Newell, S. Y. and R. R. Christian. 1981. Frequency of dividing cells as an estimator of bacterial productivity. *Appl. Environ. Microbiol.* 42: 23–31.
- Payne, W. J. 1970. Energy yield and growth of heterotrophs. *Ann. Rev. Microbiol.* 24: 17–52.
- Pomeroy, L. R. 1974. The ocean's food web, a changing paradigm. *BioScience* 24: 499–504.
- and R. G. Wiegert. 1981. The Ecology of a Salt Marsh. Springer-Verlag, New York. 271 pp.
- , W. M. Darley, E. L. Dunn, J. L. Gallagher, E. B. Haines and D. M. Whitney. 1981. Primary production. Pages 29–68 in L. R. Pomeroy and R. G. Wiegert, eds. *The ecology of a salt marsh*. Springer-Verlag, New York.
- Ragotzkie, R. A. and R. A. Bryson. 1955. Hydrography of the Duplin River, Sapelo Island, Georgia. *Bull. Mar. Sci.* 5: 297–314.
- Rassoulzadegan, F. 1982. Dependence of grazing rate, gross growth efficiency and food size range on temperature in a pelagic oligotrichous ciliate *Lohmanniella spiralis* Leeg., fed on naturally occurring particulate matter. *Ann. Inst. Oceanogr. Paris* 58: 177–184.
- Robertson, J. R. 1983. Predation by estuarine zooplankton on tintinnid ciliates. *Est. Coastal Shelf Sci.* 16: 27–36.
- Sherr, B. F. and E. B. Sherr. 1983. Enumeration of heterotrophic microprotozoa by epifluorescence microscopy. *Est. Coastal Shelf Sci.* 16: 1–7.
- , and T. Berman. 1983. Grazing, growth and ammonium excretion rates of a heterotrophic microflagellate fed with four species of bacteria. *Appl. Environ. Microbiol.* 45: 1196–1201.
- Sieburth, J. McN. and P. G. Davis. 1982. The role of heterotrophic nanoplankton in the grazing and nurturing of planktonic bacteria in the Sargasso and Caribbean seas. *Ann. Inst. Oceanogr. Paris* 58: 285–296.
- Sottile, W. S., II. 1973. Studies of microbial production and utilization of dissolved organic carbon in a Georgia salt marsh estuarine ecosystem. Ph.D. Dissertation, University of Georgia, Athens. 165 pp.
- Thomas, J. P. 1971. Release of dissolved organic matter from natural populations of marine phytoplankton. *Mar. Biol.* 11: 311–323.

- Wetzel, R. L. and R. G. Wiegert. 1983. Ecosystem simulation models: tools for the investigation and analysis of nitrogen dynamics in coastal and marine ecosystems. Pages 869–892 in E. J. Carpenter and D. G. Capone, eds. Nitrogen in the marine environment. Academic Press, New York.
- Wiebe, W. J. and L. R. Pomeroy. 1972. Microorganisms and their association with aggregates and detritus in the sea: a microscopic study. Mem. Ist. Ital. Idrobiol. Suppl. 29: 325–352.
- Wiegert, R. G. 1973. A general ecological model and its use in simulating algal-fly energetics in a thermal spring community. Pages 85–102 in P. W. Geier, L. R. Clark, D. J. Anderson and H. A. Nix, eds. Insects: Studies in population management. Vol. 1. Occasional Papers, Ecol. Soc. Australia, Canberra.
- . 1975. Simulation modeling of the algal-fly components of a thermal ecosystem: effects of spatial heterogeneity, time delays and model condensation. Pages 157–181 in B. C. Patten, ed. Systems analysis and simulation in ecology. Academic Press, New York.
- and R. L. Wetzel. 1974. The effect of numerical integration technique on the simulation of carbon flow in a Georgia salt marsh. Proc. Summer Computer Simulation Conf. Houston. Vol. 2: 275–277.

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