toxic for EAC1–3 cells. In contrast, purified lymphocytes (*19*) exhibited a strikingly reduced cytotoxicity. These data would argue for the monocyte being the cell type responsible for the observed cytotoxic effect. However, the participation of lymphocytes in the cytotoxic reaction should not be excluded at this stage of our investigations, particularly in view of the observation (*20*) that lymphocyte transformation induced by antigen or by "mixed culture" conditions was strongly diminished when purified lymphocytes were used. It is conceivable that the cytotoxic capacity of unfractionated leukocyte preparations was due mainly to lymphocytes which were triggered into action by a small number of other leukocytes. The monocyte-rich preparations contained up to 30 percent small lymphocytes and the unfractionated leukocytes were usually more potent than the purified cell preparations.

Figure 1B illustrates the effect of lymphocyte-rich leukocytes on EAC1–7 cells. The leukocytes came from the same batch of cells used for the experiment represented in Fig. 1A. Almost all cells were lysed within 4 hours. Thus, kinetically, this reaction is different from the usual lymphocyte-mediated cytotoxic reactions. The target cells were more labile than EAC1–3 cells, since they lost 30 percent of their isotope upon prolonged incubation even in the absence of leukocytes. However, the lability of EAC1–7 cells was not a constant phenomenon; the isotope release in leukocyte-free controls varied from 8 to 30 percent within 14 hours. Pretreatment of leukocytes with antimycin A led to a strong reduction of lysis (Fig. 1B). EAC1–7 cells were efficiently lysed both by glass bead purified lymphocytes (*19*) and by the monocyte-enriched preparations (*18*). Thus, lymphocytes, and perhaps also monocytes, appear to be effector cells in this cytotoxic reaction. The effect of these cells on EAC1–7 resembles that of C8, which is capable of lysing EAC1–7 cells in the absence of C9. Characteristically, this reaction is considerably slower than that mediated by C8 and C9. One hypothesis, which is presently being investigated, proposes that lymphocytes and perhaps some other leukocytes lyse EAC1–7 cells by releasing C8 or C8 and C9. This assumption finds some support in results obtained with leukocytes pretreated with rabbit antiserum to purified human C8 (*21*). This treatment inhibited their phytohemagglutinin-induced cytotoxic effect on complement- and antibody-free chicken erythrocytes. Rabbit antiserum to human C1q, C2, C3, or C4 had no such effects, while anti-C5 had a weak inhibitory effect.

Regardless of the exact mechanism underlying our observations, we conclude that complement may participate in cell-mediated cytotoxic reactions. The results underline the importance of the complement system as a link between humoral and cellular immune reactions of a tissue-damaging nature.

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Green Sea Turtles: A Discrete Simulation of Density-Dependent Population Regulation

Abstract. *Field data on the nesting of the green sea turtle were used to construct a stochastic model. This model was simulated by use of a digital computer language simscript (Monte Carlo) to determine the relation between the percentage of nests destroyed and the size of the turtle population. Nest destruction is dependent on population density and provides a mechanism to regulate population size.*

During a study on the ecology of the green sea turtle *Chelonia mydas* (L.) on the Great Barrier Reef we observed that, where the density of the population was high, the nesting turtles frequently destroyed their eggs. All turtles were tagged, and we kept a complete record of the movement of every turtle for 5 weeks. Furthermore, every nesting turtle was observed over a 14-week period in each of four successive years. These data formed the basis for constructing a stochastic model.

Turtles come ashore after dark, usually around high tide, and nest in a narrow zone above the spring high-tide mark in the outer limits of the vegetation. Turtles first dig a body pit about

1.5 m by 1.2 m by 45 cm deep; in the back of this a round egg chamber, with a cross section of approximately 30 cm, is dug about 40 cm deeper. Many of these excavations cave in during construction and are abandoned (*1*), an average of 2.4 being dug to lay a clutch of eggs. The turtle population studied laid up to four clutches of eggs at approximately 15-day intervals. After completing a nesting season, turtles do not return to breed for at least 4 years. Thus several discrete populations use the same nesting beach in different years.

The following information was used in the simulation: distribution of the arrival at the start of the season; number of returns for nesting or attempting to nest made by each; number of days between returns; distribution of digging activity for 35 nights; area (0.1 m^2) occupied by an egg chamber; mean number of egg chambers dug before a clutch of eggs was successfully deposited; and nesting pattern on this area (approximated by a triangular distribution at right angles to the beach and a uniform distribution parallel to the beach).

We simulated the observed nesting behavior of this population in order to ascertain the relation between the percentage of nests destroyed and size of the population. This model was constructed to find out whether the increasing destruction of the eggs occurred as the density of the population increased, so that, as a result, the size of the population remained relatively stable.

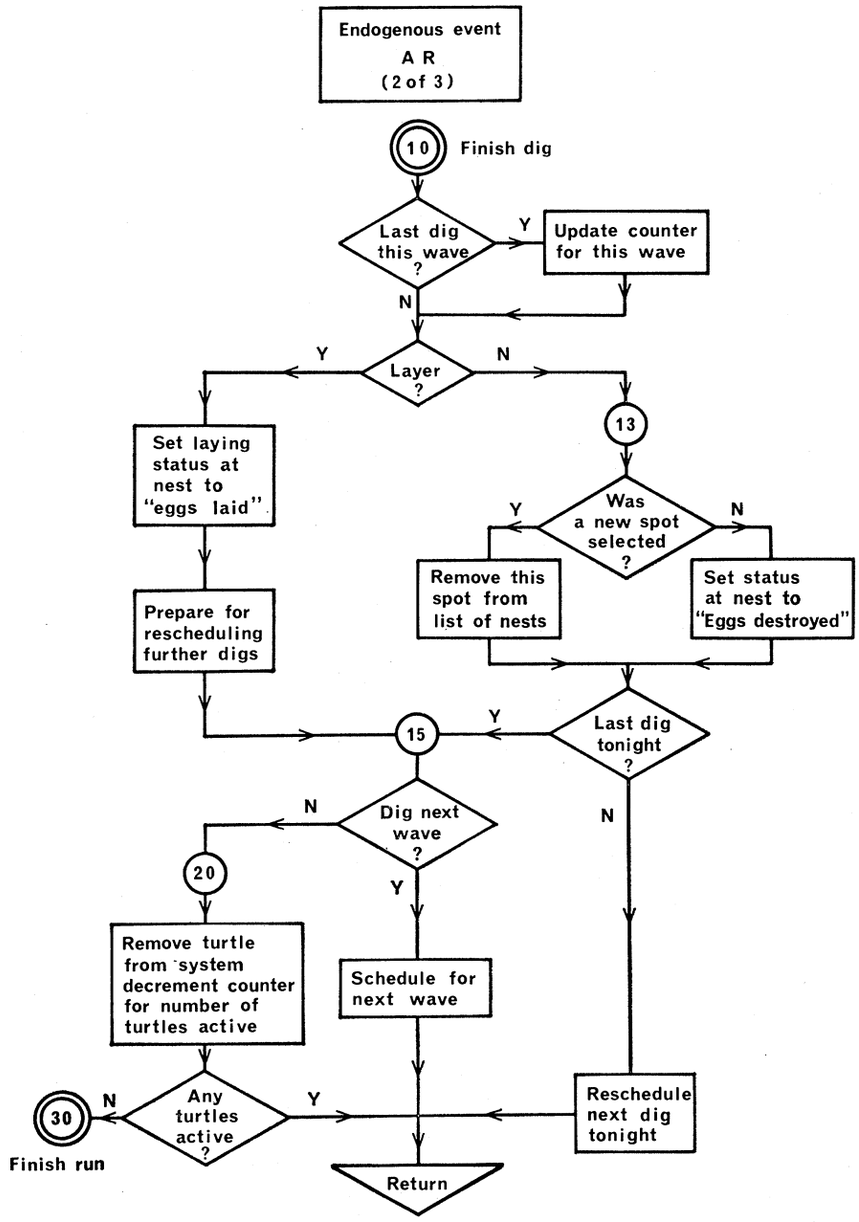

Fig. 1. Portion of flow chart indicating programming detail for completion of digging.

This oversimplified form of the model elucidates the structure of the more general model. If on any night, each turtle digs once and lays, and the turtles select different sites from a uniform distribution, the process for any night will be equivalent to sampling without replacement, and thus the distribution of nests destroyed will be hypergeometric. Thus, probability (k_n nests destroyed on nth night) is

$$\binom{r_n}{k_n}\left[\frac{s-r_n}{j-k_n}\right]\Big/\binom{s}{j_n}$$

where s is number of sites on beach available for nesting; r_n is number of nests containing eggs on nth night; j is number of turtle arrivals per night.

Over n nights the model shows a rather complex branching process, and the distribution of the fraction of nests destroyed cannot be simply represented analytically. However on the nth night, the expected number of nests destroyed is $r_n p$ (where $p = j/s$). Considering only expectations one can show that the expected fraction of nests destroyed is

$$1 - [(1 - q^n)/(pn)] \sim p(n-1)\ /\ 2$$

where $q = 1 - p$). Thus the expected percentage of nests destroyed after many nights is directly proportional to the turtle population. This result is used as a check on the more general model.

This simple model is simulated to obtain the distribution of the fraction of nests destroyed. In a bag of s white balls, j balls are replaced with black balls; these represent the number of nests after the first night. A sample of j balls is removed from the bag, and the number of black balls in the sample is counted. This is equal to k_2; j black balls are put back into the bag, and this represents the nesting state of the beach before the arrival of turtles on the third night. This process is repeated $n - 2$ times. Then the fraction of nests destroyed is

$$\sum_{i=2}^{n} k_i/nj$$

By repeating the whole process many times the required distribution function for the fraction of nests destroyed is obtained.

If the simple model is extended to the more general model in which, for instance, j is highly variable, the number of abortive digs is random, the selection

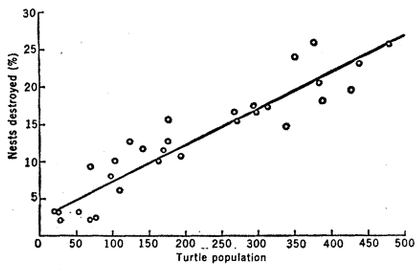

Fig. 2. Correlation of nest destruction with size of turtle population.

procedure is nonrandom, the complexity of the stochastic process is increased, and analytical and hand-simulation techniques become inapplicable. The only practical technique is a discrete computer simulation (in effect a general-purpose Monte Carlo procedure). The most powerful technique is Simscript. For this simulation, the digging of the turtles was described in extremely fine detail (Fig. 1).

We conducted a series of simulations over a range of population sizes encompassing and extending the field situation. A linear regression (Fig. 2), which fits these results with an associated probability of less than .001, indicates that nest destruction by turtles is density-dependent.

For example, we may assume that a stable turtle population contains 50 breeding females each having laid 100 eggs, and that mortality is constant, and at a level such that one female out of 95 eggs will survive to breed. Then a population with self-induced mortality (by nest destruction) of 5 percent (Fig. 2) would have 95 hatchlings per female, and the rate of population increase (P) would be 1.0, giving the same number of females in the next generation. However, if there were 300 breeding females, self-induced mortality would be 17 percent, hatchlings per female 83, and $P = 83/95 = 0.87$. The next generation would be about 261 females, that is to say population would decrease. At a population of 500 breeding females, the next generation would be only 385. In both instances decreases would continue to occur in succeeding generations, until P approaches 1.

The mechanism, therefore, tends to keep a population within certain limits. At low population density its effects are negligible; however, if a population increased greatly it would not stay at the new level due to this mechanism, which tends to restore the population to its original level.

Variation is the important attribute of mortality (*2*), low but variable mortalities having the same influence on population trends as high but relatively constant mortalities. Most factors in mortality appear to be density-dependent, and variations at high levels are potentially more important. Effects of one or a few ecological processes may account for most of the variability in trend in population numbers (*3*).

There are no nest predators on Barrier Reef cays, and the green turtle has been entirely protected by law since 1950; hence we are dealing with large natural populations of this turtle. However, predation (both human and nonhuman) is very high on many other beaches where turtles nest (*4, 5*). We anticipate that, where the species has been seriously overexploited, the population density is depressed below the level at which nest destruction by turtles would operate as a regulatory mechanism. The Barrier Reef of Australia may be the last place where natural regulation of population size by the mechanism of density-dependent nest destruction can still be observed today.

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Mitotic Division in Pancreatic Beta Cells

Abstract. *Successful expansion of the islet cell mass occurs in genetically diabetic mice (C57 Bl/Ks-dbdb) following a period of dietary restriction, in the absence of a population of precursor cells. Differentiated cells that synthesize insulin retain the capability of undergoing mitotic division.*

The pancreatic diverticulum develops as an evagination of the primitive gut, and, in the subsequent morphogenesis of the embryo, is transformed into the ducts and acinar cells of the exocrine pancreas (*1*). The belief that the forerunners of the islets of Langerhans also originate in epithelial cells of the primitive ducts (*1*) has recently been questioned. Noting that mesodermal cells accumulate in juxtaposition to the evaginating pancreatic diverticulum, Wessels (*2*) was unable to determine whether the initial cells possessing beta granules were ductal (endodermal) or mesodermal in origin. The frequent observation of proliferating ductal elements in the pancreases of diabetic experimental animals has been interpreted as evidence of postnatal beta cell neogenesis (*3*). However, convincing ultrastructural evidence of transformation of duct cell to islet cell is lacking. The conversion of pancreatic acinar cells into islet cells was originally suggested (*4*) as a physiologic means of varying the relative exocrine and endocrine functional capacities. This hypothesis has few modern advocates (*5*) and has suffered from the absence of acceptable ultrastructural evidence of transition forms between the two cell types. Mitotic division among islet cells has not been stressed as a significant cause of postnatal beta cell proliferation because mitoses are not frequently observed among islets; furthermore, it is rarely, if ever, possible to determine with the light microscope the cell type undergoing mitosis. One may question, as workers using other systems have (*6*), whether cells that synthesize a protein such as insulin are too differentiated to undergo cell division.

We find that insulin-producing cells do divide and suggest that mitotic division among differentiated beta cells may be an important mode of their postnatal proliferation.

Diabetic mutants (dbdb) of C57 Bl/Ks mice, if allowed free access to food, die after 5 to 7 months with evidence of marked hyperglycemia and decreasing concentrations of insulin in the serum (*7*). Their islets of Langerhans reveal decreased numbers of beta cells and numerous small ducts (Fig. 1). The latter have been interpreted as evidence of an unsuccessful stimulation of beta cell neogenesis (*7*). When food is made available for only 8 hours per day on Monday, Wednesday, and Friday of