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AUTHOR(S): Ramit Mehr, CNLS
Zvia Agur, Weizmann Institute of Science

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TEMPORAL STOCHASTICITY LEADS TO NONDETERMINISTIC CHAOS IN A MODEL FOR BLOOD CELL PRODUCTION

Ramit Mehr[a,*] and Zvia Agur[b]

Department of Applied Mathematics
The Weizmann Institute of Science
Rehovot 76100, Israel
[a]Present Address: Center for Nonlinear Studies (CNLS)
Mail Stop Box 258
Los Alamos National Laboratory (LANL)
Los Alamos, NM 87545
[b]Present Address: Department of Zoology
and Centre for Infectious Diseases and Epidemiology
University of Oxford
South Parks Road, OX1 3PW, Oxford, UK

* *To whom correspondence should be addressed*

ABSTRACT

All types of blood cells are formed by differentiation from a small population of pluripotent stem cells in the bone marrow. This population should maintain the balance between self-renewal and differentiation, even under severe perturbations, e.g. the massive cell death caused by chemotherapy or irradiation. We constructed a cellular-automata model for bone marrow dynamics, which retrieves its homeostatic capabilities even under periodic perturbations with constant or random amplitude. However, temporally stochastic perturbations result in a chaotic-like behaviour. Several methods of analysis failed to distinguish between the time series in this case and a chaotic time series, although the chaotic-like behaviour has no deterministic source.

I. INTRODUCTION

In the process of hemopoiesis, a small population of pluripotent stem cells continuously produces the whole variety of mature blood cells, while maintaining the stem cell pool through self-renewal. The hemopoietic system is a good example of a homeostatic, self-organizing, biological system. The probabilities of self-renewal, i.e., cell divisions that generate daughter cells identical to the mother cell, and of differentiation of daughter cells into blood cell precursors, are adjusted according to the demands of the organism. The balance between self-renewal and differentiation is maintained even under harsh cytodestructive (cell killing) treatments, e.g., chemotherapy or irradiation. Some of the relevant features of this system are reviewed in the next section.

In a previous study [1] we used a cellular automata model of hemopoiesis to study the dynamics of blood cell production under various regimens of treatment. Our model showed an ability to maintain homeostasis under periodic treatment regimens, even when the dose administered was random. However, chaotic-like behaviour was observed when we introduced stochasticity in the time intervals between treatment "perturbations". These results are reviewed in section III.

The chaotic-like behaviour observed in our simulations has a significance that is not limited to models of blood cell production. Lately there has been a multitude of publications announcing the observations of chaotic behaviour in various biological systems. Research efforts have been directed to explaining why chaos should be an essential part of the dynamics of heart or brain function (reviewed in section IV). However, our results - showing a behaviour that is indistinguishable from chaos on the basis of several statistics, in spite of the fact that this "chaos" is generated by temporal noise alone - suggest that stochastic driving may be the cause of the "chaos" observed in some of the other cases as well.

II. THE DYNAMICS OF BLOOD CELL PRODUCTION

Bone-marrow stem cells give rise to precursor cells that are irreversibly committed to differentiate into one of the various hemopoietic lineages. Committed cells further differentiate into immature, lineage-restricted cells that mature into eight main types of specialized cells. Proliferation and differentiation are controlled by protein signals (cytokines) that are secreted by hemopoietic cells themselves, by bone marrow stromal cells, and by other organs [2, 3]. The spatial structure of the bone marrow plays a role in the control process: a neighbourhood of cells committed to differentiation seems to favor differentiation, while a stem-cell neighbourhood favors self-renewal [4]. This poorly understood feedback [5–7] modulates the balance between the number of stem cells that differentiate and those that self-renew, ensuring the existence of required numbers of any cell type [8]. Considerable research efforts have recently been directed towards the identification of the most primitive hemopoietic stem cell, which is possibly the one bearing the CD34 surface antigen [9–12], and assessing its self-renewal capacity [13, 14]. The most

studied, though probably not the most primitive, are the spleen colony-forming units (CFU-S). In a steady state, 90% of these cells are proliferatively quiescent [15, 16], but after a severe depletion, which may be caused by chemotherapy or irradiation, as few as one or two dozens of CFU-S suffice for complete regeneration of bone marrow and blood cells [6, 17].

Homeostasis of bone marrow cell populations, in particular the stem cell population, is essential for the normal function of the organism. When this homeostasis is not maintained, as in some pathological situations, various hemopoietic and immune disorders may occur. One specific type of these disorders was termed "dynamical diseases" [18]. An example of this type of diseases is cyclic neutropenia – a condition in which the levels of neutrophils (one of the white blood cell types) in the blood exhibits large fluctuations over time [19, 20].

How the bone marrow homeostasis is maintained and under which conditions it is upset are questions of major importance, especially now when drug therapy is considered in conjunction with bone marrow reconstitution [21]. Drug therapy or irradiation are used in a variety of blood cell and other malignancies. An effective treatment should not only maximize the damage to malignant cells but also minimize the damage to the patient's normal cells [22–28]. In addition, when bone marrow transplantation is considered as a means of restoring the patient's hemopoietic and immune systems, drugs and irradiation are used prior to transplantation to eradicate the patient's diseased cells. Hence the most efficient eradication regimen should be used [21, 29].

III. A COMPUTER MODEL OF STEM CELL DYNAMICS

The aim of the present work is to study the property of homeostasis that characterizes the bone marrow, and to analyze the effect of different drug regimens on its regeneration ability. The rationale of our mathematical modelling is to find the minimal assumptions necessary to retrieve the observed phenomenon. Accordingly, we assumed that each cell may be in one of three possible states: one is the stem cell, and the two other states are cells already committed to one of two branches of differentiation. A "stem cell" in our model represents the most primitive stem cell, which has a high capacity (infinite, in the model) of self-renewal (the exact identity of the stem cell is irrelevant). A "stem cell" in the model switches from self-renewal into differentiation according to the states of its neighbouring cells. Thus, the balance between self-renewal and differentiation is controlled only through communication between neighbouring cells, while proliferation is controlled only by available space. We assumed that additional factors which are involved in the global control of the system, and not in the local cell-cell interactions, are of secondary importance relative to local interactions [7]. For this reason, in our model we did not include such factors - again striving to construct a minimal description. Inter-cell interactions in real bone-marrow may be stimulatory [14, 30–33] or inhibitory [6, 7, 34–37]. However, the existence of growth

inhibitors in the bone marrow is as yet to be fully understood. No specific inhibition on stem-cell proliferation or differentiation is assumed in our model: the stem cell tends to "decide" its next move according to the principle that an environment composed mainly of stem cells encourages self-renewal, and an environment of more mature cells, committed to differentiation, encourages differentiation. We did not argue that there is no inhibition, but, rather, we examined the possibility that the basic properties of hemopoiesis can be retrieved without assuming inhibition.

The above assumptions were realized in a cellular automata model [1], consisting of a two-dimensional lattice containing 10,000 cells. Each automaton cell represented a real-life stem cell or differentiated cell in the bone marrow. Initially, the automata are populated by 10,000 stem cells, and those cells then evolve in time according to local rules. Each simulation step corresponds to a cell division cycle, so that time is measured in multiples of the cycle time of bone marrow cells. The maximum lifetime of a differentiated cell in the bone marrow is τ, meaning that a differentiated cell dies τ simulation steps after it was generated by differentiation from a stem cell. Thus τ turns out to be the characteristic time scale of our system. As long as the system is unperturbed, and its differentiation rate is not larger than the stem cells' self-renewal rate, it evolves to a steady state where the proportion of the various cell types in the population stays roughly the same. If the differentiation rate exceeds the stem cells' self-renewal rate, the pool of stem cells is exhausted and all cells eventually "die". The results of the simulations of our model are reviewed in the following. We present the dynamics in terms of one variable, s, which is the fraction of stem cells (out of the maximum possible number of cells) in the simulated population. For the system to be able to return to its steady state after perturbations, this fraction should not go down to near-exhaustion levels.

Homeostasis was exhibited in our model by a full recovery from severe (up to 97%) stem-cell loss. The number of stem cells, s, exponentially recovers towards the steady state, which we denote by $\hat{s}$. On this exponential recovery, damped oscillations of period τ are superimposed. These oscillations were generated by the synchronization of cell death and proliferation in a large fraction of the population, occuring after a single sharp depletion. Similar waves of synchronized proliferation were observed in bone marrow stem cells of mice, and explained as the result of a hypothetical autoregulation of stem cells through a proliferation-inhibiting factor [38, 39]. Our work suggests that there need be no such factor, since the synchronized cell death following each perturbation can in itself result in such oscillations.

Simulating various regimens of toxic-drug administration by repeated depletion "perturbations", we have shown that homeostasis can be maintained when drug administration is fully periodic (Fig. 1). In contrast, when time intervals between administrations become stochastic, chaotic-like behaviour emerges (Fig.

2), and the stem-cell pool is eventually depleted. The temporal behaviour of the stochastically-perturbed system was highly sensitive to initial conditions: a change in the state of as little as 0.001 of the cells in the initial configuration results in totally different time-series. Our system also exhibits almost complete memory loss, measured by the Gade-Amritkar method [40]: $\frac{\nu_{(t>0)}}{\nu_{(t=0)}}=2.0 \pm 0.1$, where doubling of the generalized exponent ν_t indicates complete memory loss, characteristic of chaotic time-series. The periodically-perturbed series exhibit only partial memory loss, which is due to intrinsic noise in the system: $\frac{\nu_{(t>0)}}{\nu_{(t=0)}}=1.5 \pm 0.1$.

We performed an analysis of this behaviour, via the time-evolution equations for the stem-cell pool. We looked only at averaged quantities (averaged over the whole grid of cells), which is equivalent to making the simplifying assumption of homogeneity. The behaviour of the average stem cell fraction turned out to be described by a logistic-type equation, with parameters in the one steady state regime [1]. To examine the source of the chaotic-like behaviour, we studied the stem-cell fraction on the instant just prior to the perturbation. Define S_j as the value of s at the instant *prior* to the j'th perturbation. Between perturbations s recovers exponentially with some characteristic time β (which can be calculated from model parameters). After each perturbation, which eliminates a fraction D of the stem cells, s equals $(1-D)S_j$. Hence S_j (which is a *Poincaré* mapping [41] of the original time series) evolves in time through:

$$S_{j+1} = [(1-D)S_j - \hat{s}]e^{-\beta t_j} + \hat{s}, \tag{1}$$

where t_j is the time between the j'th perturbation and the (j+1) perturbation.

In the *periodically* perturbed case, $t_j = T$ = constant. The mapping is linear and as such has only one (stable) fixed point:

$$\hat{S} = \frac{1-e^{-\beta T}}{1-(1-D)e^{-\beta T}}\hat{s} \tag{2}$$

This resembles the "mode locking" phenomenon characteristic of periodically-driven systems, appearing also in neural network models [42, 43].

In the chaotic-like case, t_j in Eq. (2) is a *random* variable, normally distributed with an average $\overline{T}$ and a standard deviation σ. The value of S_j, which depends on t_j, may fluctuate faster than the system can adjust, i.e. the perturbation "hits" at a different point of the recovery to equilibrium every time, so that the system is unable to settle onto periodic dynamics (unless $\overline{T}$ is so large - of the order of 200 simulation steps - that the system returns to the steady state before each perturbation). This noise in t_j can never drive the stem cell fraction to the chaotic regime, because the calculations are performed only in the one fixed-point regime. The source of the chaotic-like behaviour we see must then lie in the

stochastic driving itself. Driven by temporally stochastic perturbations, the system constantly "jumps" between periodic orbits with different periods, and this results in a behaviour that looks chaotic. Thus, these stochastic perturbations destabilize the system in a way comparable to the case described by Newhouse [44–46], where there are many complex attracting periodic orbits of arbitrarily high periods, with very narrow and convoluted domains of attraction, so that any amount of noise leads to chaotic behaviour.

IV. CHAOS IN BIOLOGICAL SYSTEMS

Low-dimensional erratic behaviour has recently been observed in several biological systems [47–50]. This was assumed to indicate that the underlying behaviour was deterministic chaos, e.g., in blood flow or heart rate [51]. In some cases, e.g., in the nervous system, it was furthermore conjectured that the chaotic mechanism plays the role of a maximal-information basic state of the systems considered [47, 52].

We suggest, based on our model for hemopoietic stem-cell dynamics, that a behaviour that looks chaotic may sometimes result from stochasticity in the intervals between successive perturbations, and is not necessarily deterministic. It is worth noting that $\frac{1}{f}$ noise does not always imply deterministic chaos, and fractal dimension measurements are also problematic as a means of distinguishing deterministic chaos from stochasticity [53].

Results presented here support previous conclusions [54, 55] that the distribution of inter-disturbance intervals is the major factor in determining population persistence. The possibility that random intervals between high-dose drug applications may impede bone-marrow regeneration should be further investigated. In this context it should be noted that our mathematical work and laboratory experiments also suggest that fully periodic drug application may minimize the cytotoxicity to the bone-marrow [22–28, 56].

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REFERENCES

[1] R. Mehr and Z. Agur, BioSystems 26, 231 (1992).

[2] M.A.S. Moore, M.O. Muench, D.J. Warren, and J. Laver, In: Molecular Control of Hemopoiesis, edited by Bock, G., and Marsh, J., (Wiley, 1990).

[3] D. Metcalf, Nature 339, 27 (1989).

[4] D. Zipori, In: Hematopoiesis, long term effects of chemotherapy and radiation, edited by N.G. Testa and R.P. Gale, (Marcel Dekker, NY-Basel, 1988), vol. 8, Ch. 2.

[5] B.I. Lord and R. Schofield, Lect. Notes in Biomath. 38, 9 (1979).

[6] D. Zipori, In: The biology of Hemopoiesis, edited by N. Dainiak, E.P. Cronkite, R. McCaffrey and R.K. Shadduck, Progress in Clinical and Biological Research series, No. 352, (Wiley-Liss, 1990). Proc. of 15th F. Stohlman Jr. Memorial Symposium, Cambridge, Mass. October 15-20, 1989. p.115.

[7] D. Zipori, Cancer Cells 2(7), 205 (1990).

[8] H.E. Wichmann, M. Loeffler and S. Schmitz, Blood Cells 14, 411 (1988).

[9] J.W.M. Visser and D.W. Van Bekkum, Exp. Hematol. 18(3), 248 (1990).

[10] I. Bertoncello, T.R. Bradley and S.M. Watt, Exp. Hematol. 19(2), 95 (1991).

[11] I. Bertoncello, T.R. Bradley, G.S. Hodgson and J.M. Dunlop, Exp. Hematol. 19(3), 174 (1991).

[12] M. Kobayashi, A., Mullbacher, P. Waring and A.J. Hapel, Eur. J. Haematol. 46(4), 205 (1991).

[13] H.J. Sutherland, P.M. Lansdorp, D.H. Henkelman et al, PNAS 87, 3584 (1990).

[14] S. Okada, T. Suda, J. Suda, N. Tokuyama, K. Nagayoshi, Y. Miura and H. Nakauchi, Exp. Hematol. 19(1), 42 (1991).

[15] B.I. Lord and N.G. Testa, In: Hematopoiesis, long term effects of chemotherapy and radiation, edited by N.G. Testa and R.P. Gale, (Marcel Dekker, NY-Basel, 1988), vol. 8, pp. 1-26.

[16] R. Schofield, Blood Cells 4, 7 (1978).

[17] G.J. Spangrude, S. Heimfeld and I.L. Weissman, Science 241, 58 (1988).

[18] L. Glass and M.C. Mackey, Ann. N.Y. Acad. Sci. 316, 214 (1979).

[19] W. P. Hammond, T.C. Boone, R.E. Donahue, L.M. Souza and D.C. Dale, Blood 76(3), 523 (1990).

[20] S. Tsunogake, S. Nagashima, R. Maekawa, N. Takano, H. Kajitani, K. Saito,

H. Enokihara, S. Furusawa and H. Shishido, International Journal of Hematology 54(3), 251 (1991).

[21] K. Atkinson, Bone Marrow Transplantation 5, 209 (1990).

[22] Z. Agur, In: Perspectives in biological dynamics and theoretical medicine. Annals of the New York Academy of Sciences 504, 274 (1986).

[23] Z. Agur, Lancet 334(ii), 734 (1989).

[24] Z. Agur, In: Biomedical modelling and simulation, edited by J. Eisenfeld and D.S. Levine, (J.C. Baltzer AG, Scientific Publishing Co., 1989), IMACS, pp 59-61.

[25] Z. Agur, R. Arnon and B. Schechter, Math. Biosc. 92, 1 (1988).

[26] Z. Agur, R. Arnon and B. Schechter, Eur. J. Cancer 28A(6/7), 1085 (1992).

[27] L. Cojocaru and Z. Agur, Math. Biosc. 109, 85 (1992).

[28] G. Webb, To appear in the Proceedings of the World Congress on Nonlinear Analysis (1993).

[29] R. Mehr, M. Fridkis-Hareli, L. Abel, L. Segel and A. Globerson, (unpublished).

[30] H.E. Broxmeyer, S. Cooper, L. Lu, G. Hangoc, D. Anderson, D. Cosman, S.D. Lyman and D.E. Williams, Blood 77(10), 2142 (1991).

[31] P.J. Quesenberry, H.E. McGrath, M.E. Williams, B.E. Robinson, D.H. Deacon, S. Clark, D. Urdal and I.K. McNiece, Exp. Hematol. 19(1), 35 (1991).

[32] I.K. McNiece, K.E. Langley and K.M. Zsebo, J. Immunol. 146(11), 3785 (1991).

[33] I.D. Bernstein, R.G. Andrews and K.M. Zsebo, Blood 77(11), 2316 (1991).

[34] G.J. Graham, E.G. Wright, D. Donaldson, S. Lorimore and I.B. Pragnell, Exp. Hematol. 18(6), 713 (1990).

[35] G.J. Graham, E.G. Wright, R. Hewick, S.D. Wolpe, N.M. Wilkie, D. Donaldson, S. Lorimore and I.B. Pragnell, Nature 344, 442-444 (1990).

[36] C.S. Johnson, S.C. Pourbohloul and P. Furmanski, Exp. Hematol. 19(2), 101 (1991).

[37] B. Bungart, M. Loeffler, H. Goris, B. Dontje, V. Diehl and W. Nijhof, Br. J. Hematol. 76(2), 174 (1990).

[38] E. Nečas and V. Znojil, Cell Tissue Kinet. 21, 73 (1988).

[39] F. Hauser and E. Nečas, Cell Tissue Kinet. 21, 81 (1988).

[40] P.M. Gade and R.E. Amritkar, Phys. Rev. Lett. 65, 389 (1990).

[41] J.P. Eckmann and D. Ruelle, Rev. Mod. Phys. 57, 617 (1985).

[42] S. Renals and R. Rohwer, J. Stat. Phys. 58, 825 (1990).

[43] H. Sompolinski, A. Crisanti and H.J. Sommers, Phys. Rev. Lett. 61, 259 (1988).

[44] S.E. Newhouse, Topology 13, 9 (1974).

[45] S.E. Newhouse, Publ. Math. IHES, 50, 101 (1979).

[46] S.E. Newhouse, in: Dynamical Systems, CIME Lectures Bressanone, Italy, June 1978. Progress in Math., No. 8. (Birkhauser-Boston, Boston, 1980), pp.1-114.

[47] A. Babloyantz and A. Destexhe, PNAS 83, 3513 (1986).

[48] C.A. Skarda and W.J. Freeman, Behav. Brain Sci. 10, 161 (1987).

[49] R. Pool, Science 243, 604 (1989).

[50] Y. Lenbury and P. Pacheenburawana, Biosystems 26(2), 117 (1991).

[51] T.A. Denton, G.A. Diamond, R.H. Helfant, S. Khan and H. Karagueuzian, American Heart Journal 120, 1419 (1990).

[52] F.C. Hoppensteadt, PNAS 86(9), 2991 (1989).

[53] L. Glass and C.P. Malta, J. Theor. Biol. 145, 217 (1990).

[54] Z. Agur, J. Theor. Biol. 112, 677 (1985).

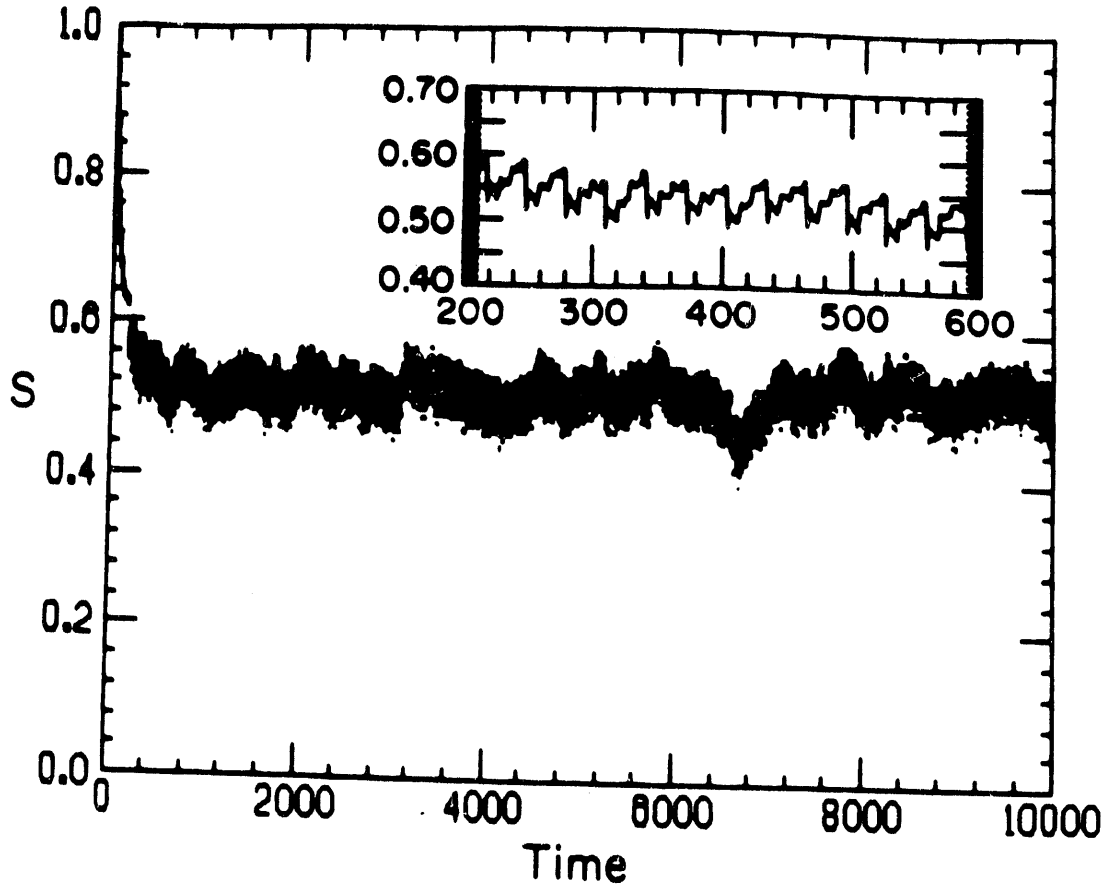
[55] Z. Agur and J.L. Deneubourg, Theor. Popul. Biol. 27, 75 (1985).

[56] Z. Agur, R. Arnon, B. Sandak and B. Schechter, Exp. Hematol. 19, 364 (1991).

FIGURE CAPTIONS

FIG. 1: The cellular automata simulations, performed on a square grid of 10^4 cells. Drug treatment is initiated after the system has reached a steady-state. Time series of the fraction of stem cells is shown here for a periodic drug treatment ($T = 30$). (Inset) Part of the series enlarged to show the periodicity. Figures are reproduced from Mehr and Agur, 1992, where the parameters of the simulations are given.

FIG. 2: Simulation of a stochastic drug treatment ($T = 30 \pm 1$) resulting in a chaotic-like time-series.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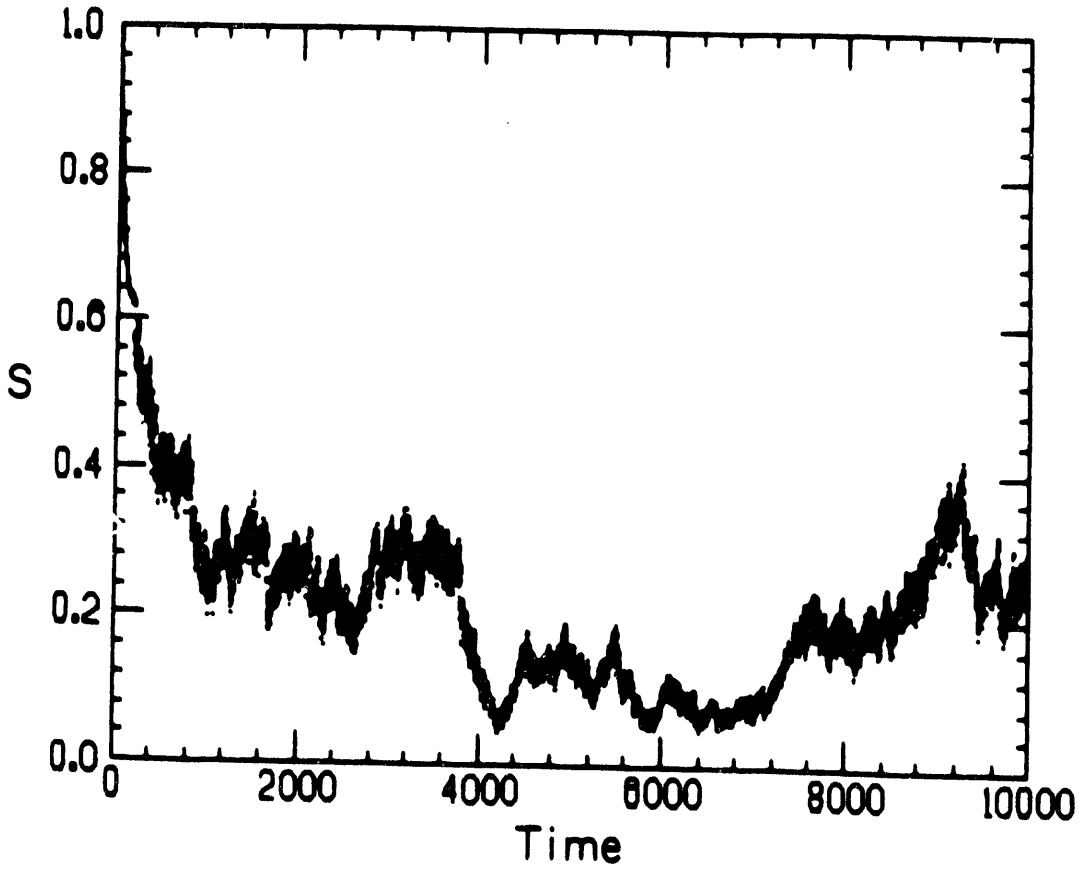
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