1

INTRODUCTION AND LITERATURE SURVEY

1.1 INTRODUCTION

Both living organisms and computers are "information-processing machines" that operate on the basis of internally stored programs, but the differences between these systems are also quite large. In the case of living organisms, self-assembly occurs following an internal program, and the nervous system and brain formed in this way function as an autonomous information machine. Unlike traditional computers which must be "driven" from the outside, biological systems have somehow incorporated within them rules on how to function. Moreover, in the case of biological entities for which there is no external blueprint, the design plan is entirely internal and is thought to undergo changes both in the evolution of species and in the development of individuals. These similarities and differences have drawn the attention of computer scientists as well as of life scientists.

In order to revolutionize the current world of computers, three roads, or any combinations of them, are clearly visible [1]

1. Changing the physical elements at the foundations of the computer components
2. Changing the architecture of computers
3. Devising new software and computing algorithms

Information Processing by Biochemical Systems: Neural Network–Type Configurations, By Orna Filo and Noah Lotan

It is, however, true that a biological computer (or *biocomputer*) of a completely different nature from today's electronic computers already exists in the form of the fundamental phenomenon of life. The most advanced machinery, a living organism, operates with functional elements that are of molecular dimensions and actually exploits the quantum-size effects of its components [1]. Yet the quintessentially biological functions of living forms: autonomy, self-organization, self-replication, and development, as witnessed in both evolution and individual ontogeny, are completely absent from current computing machines [1].

Two major approaches to the construction of a biocomputer are reviewed here:

1. Study of the operational mechanism of biological systems, particularly those of the living brain, and the use of these results in the redesign of computer software and hardware architecture based on semiconductor technology (Section 1.2).
2. Development of biocomponents that are similar to and/or composed of biological macromolecules, the development of biochips that make use of these components, and ultimately, the construction of biocomputers (Section 1.3).

1.2 COMPUTATIONAL PROCESSES BASED ON BIOLOGICAL PRINCIPLES

1.2.1 Modeling Biological Processes

The involvement of biology might lead to new computational processes based on those found in natural systems. Multiple modes of processing contribute to the information-processing functions of biological systems, and these have been investigated and modeled extensively [2–8]. In his pioneering work, Rosen [9,10] introduced a two-factor model based on the idea that the fundamental dynamic behavior of physiological and biochemical systems is regulated by the combined action of two factors, one excitatory and the other inhibitory. Kampfner, Kirby, and Conrad [11–13] introduced theoretical models of enzymic neuron systems for learning processes, based on the concept of a hypothetic enzyme called excitase. Based on the same concept, a comprehensive mathematical model of the enzymic neuron as a logical circuit has been introduced by Neuschl and Menhart [14].

1.2.2 Artificial Neural Networks

The nerve cell has proved to be an extremely valuable source of ideas about networks of automata. A fundamentally different approach to computation

is represented by artificial neural networks (ANNs), which are designed to mimic the basic organizational features of biological nervous systems [15–22]. The building brick of any neural computing system is some sort of representation of the fundamental cell of the brain: the neuron. Thus, ANNs consist of a large number of simple interconnected processing elements which are simplified models of neurons, and the interconnections between the processing elements are simplified models of the synapses between neurons. The processing of information in such networks occurs in parallel and is distributed throughout each unit composing the network [15–22].

There has been a steady development of neuronal analogs over the past 50 years. An important early model was proposed in 1943 by McCulloch and Pitts [23]. They described the neuron as a logical processing unit, and the influence of their model set the mathematical tone of what is being done today. Adaption or learning is a major focus of neural net research. The development of a learning rule that could be used for neural models was pioneered by Hebb, who proposed the famous Hebbian model for synaptic modification [24]. Since then, many alternative quantitative interpretations of synaptic modification have been developed [15–22].

There is no universally accepted definition of an artificial neural network. However, some definitions can be found in the literature, and examples are cited here.

- Robert Hecht-Nielsen, the inventor of one of the first commercial neurocomputers, defined [17] a *neural network* as "a computing system made up of a number of simple, highly interconnected processing elements, which process information by its dynamic state response to external inputs."
- According to the *DARPA Neural Network Study* [18]: "A neural network is a system composed of many simple processing elements operating in parallel whose function is determined by network structure, connection strengths, and the processing performed at computing elements or nodes."
- According to Aleksander and Morton [19], *neural computing* can be defined as "the study of networks of adaptable nodes which, through a process of learning from task examples, store experiential knowledge and make it available for use."
- According to Zurada [20], artificial neural systems, or neural networks, are "physical cellular systems which can acquire, store, and utilize experiential knowledge."
- According to Nigrin [21], "a neural network is a circuit composed of a very large number of simple processing elements that are neurally based. Each element operates only on local information. Furthermore

each element operates asynchronously; thus, there is no overall system clock."

- Haykin [22] offers a definition based on Aleksander and Morton [19]: "A neural network is a massively parallel distributed processor that has a natural propensity for storing experiential knowledge and making it available for use. It resembles the brain in two respects:
  - Knowledge is acquired by the network through a learning process.
  - Interneuron connection strengths known as synaptic weights are used to store the knowledge."

Significant progress in neural network research has been made in recent decades [15–22,25]. Presently, the neural network strategy is implemented at either the software or hardware level. The VLSI (very large scale integration) version of neural network implementation is a technology that has approached a certain degree of maturity [22]. Although the VLSI version serves as an impressive demonstration of the power of the new computer architecture of neural networks, it falls short of a radical design departure that is capable of capturing the structural and functional flexibility inherent in biosystems [25]. Many experts believe that neural network technology will be more robust and more powerful when its implementation becomes possible in a molecular-based "hardware" environment [25].

1.3 MOLECULAR AND BIOMOLECULAR ELECTRONICS

1.3.1 Motivation

The high-technology revolution that made the personal computer standard equipment was fueled primarily by astonishing advances in microelectronics that allow more and more circuit elements to be packed into a small integrated circuit (IC). The number of device components packaged into a single IC has grown exponentially with the passage of time [25–28]. Moreover, we witness increasing capability of each IC, increasing speed of operation, reduced consumption of energy, reduction in sizes and weights of the finished products, and reduced prices. Will this trend continue so that the device size eventually reaches the atomic scale? To many experts the answer is "not if using conventional microelectronics technology," which exploits mainly macroscopic properties of inorganic materials, because the ensuing quantum size and the thermal effects will make such devices unreliable [25,28]. Thus, today, the miniaturization and integration of electronic devices are being pushed to their physical limits [25–28].

1.3.2 Molecular Electronics

Molecular electronics is defined broadly as the encoding, manipulation, and retrieval of information at a molecular or macromolecular level [25–29]. This approach contrasts with current techniques, in which these functions are accomplished via lithographic manipulations of bulk materials to generate integrated circuits [28]. A key advantage of the molecular approach is the ability to design and fabricate devices from the bottom-up, on an atom-by-atom basis. Lithography can never provide the level of control available through organic synthesis or genetic engineering [28]. The molecular primitives allow for improvement in a number of information-processing device characteristics compared with similar characteristics of silicon-based devices. Thus, molecular information processing is attractive because it offers [29]:

- Integrability at the atomic scale
- High computational speed due to parallel processing, which compensates for the inherent low processing rate of each elementary device
- Self-assembly capability of atomic or molecular processors
- Plasticity of the molecular circuit, which can reconfigure itself to optimize its performance, taking into account the previous experience (learning)
- Fault-tolerance capability and even self-repair ability of the molecular circuit
- Reduced power consumption

Since Aviram's proposal of a molecular rectifier [30,31], a variety of designs of molecular electronic devices have appeared. Molecular-scale devices are fabricated on the nanometer scale and are composed of either a single molecule or several molecules configured into a supramolecular complex. Among these devices, molecular rectifiers, molecular switches, molecular diodes, molecular photodiodes, and molecular memories are described [30–39]. Studies also deal with assembling the individual components in thin-film configurations [25,40,41], forming artificial membranes [25,42] and establishing an interface between the molecules and conventional electronic materials [43]. Another possibility that has been investigated is the use of electroconductive polymers as "molecular wires" for establishing the connection required between molecular elements [43,44].

1.3.3 Biomolecular Electronics

Biomolecular electronics is a subfield of molecular electronics that considers the use of native and modified biological molecules in electronic or photonic

devices [45–56]. The growing interest in the possibility of utilizing biological molecules in molecular electronics is fostered by the basic understanding that, in so doing, one may be able to take advantage of the specific characteristics and unique capabilities of these natural molecules [44–56]. Among the biomolecular devices investigated, protein-based molecular devices have gained increasing attention due to the versatile and highly specific molecular functionality of proteins [43,57]. Enzymes [44,58–67], receptors [68], antibodies [43], and bacteriorhodopsin [29,69–73] have been used as either electronic or optical devices. Computation with simple DNA manipulations has also been demonstrated [74,75].

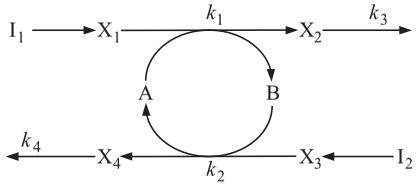
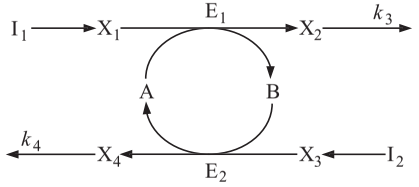
1.4 BIOCHEMICAL DEVICES BASED ON ENZYMIC REACTIONS

In an extensive study, Okamoto and co-workers [76–86] introduced a biochemical switching device based on a cyclic enzyme system in which two enzymes share two cofactors in a cyclic manner. Cyclic enzyme systems have been used as biochemical amplifiers to improve the sensitivity of enzymatic analysis [87–89], and subsequently, this technique was introduced into biosensors [90–93]. In addition, cyclic enzyme systems were also widely employed in enzymic reactors, in cases where cofactor regeneration is required [94–107]. Using computer simulations, Okamoto and associates [77,80–83] investigated the characteristics of the cyclic enzyme system as a switching device, and their main model characteristics and simulation results are detailed in Table 1.1, as is a similar cyclic enzyme system introduced by Hjelmfelt et al. [109,116], which can be used as a logic element.

Subsequently, Okamoto and associates [84–86] investigated the connection of several cyclic enzyme systems in order to construct a network. In their models the cyclic enzyme system represents a biochemical neuron that participates in a biochemical neural network. These models are detailed in Table 1.2. Theoretical models of such networks were also proposed by Hjelmfelt and co-workers [109–111,116], and these are also presented in Table 1.2.

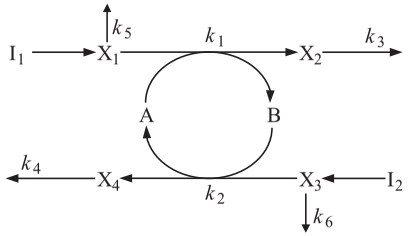
Models for biochemical switches, logic gates, and information-processing devices that are also based on enzymic reactions but do not use the cyclic enzyme system were also introduced [76,115,117–122]. Examples of these models are presented in Table 1.3. It should also be mentioned that in other studies [108,112–114,116], models of chemical neurons and chemical neural networks based on nonenzymic chemical reactions were also introduced.

Table 1.1 Models Based on the Cyclic Enzyme System

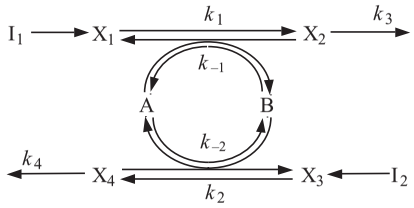
Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
1	 I_1,I_2: inputs to the system's substrate pools of X_1 and X_3, respectively. Simple mass action kinetics. Irreversible reactions.	The steady-state concentrations of A and B ($\overline{A}$, $\overline{B}$) change stepwise at $I_2/I_1 = 1$ and can be represented as $\overline{A} = f(I_1, I_2)$ $= [1 \text{ if } I_1 \geq I_2;\ 0 \text{ if } I_1 < I_2]$ The steady-state concentrations of X_2 and X_4 ($\overline{X}_2$, $\overline{X}_4$) are also a function of I_1 and I_2: $\overline{X}_2 = \overline{X}_4 = f(\min(I_2, I_2))$ $= \left[\frac{I_2}{k} \text{ if } I_1 \geq I_2;\ \frac{I_1}{k} \text{ if } I_1 < I_2\right]$ where k indicates the rate constant of the decay of X_2 or X_4, k_3 or k_4, respectively.		The mathematical equations for this model and the following ones agree with the case of batch reactions, in which there is no mass flow into or out of the system. However, I_1 and I_2 cannot be defined as mass flows if volume changes are not considered.	77
2	 Same assumptions as in model 1 except: Reaction mechanisms are assumed to be ordered bi–bi enzymic reactions.	Same results for X_2 and X_4 as obtained in model 1.			77

(*continued*)

Table 1.1 (*continued*)

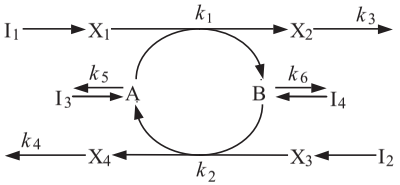
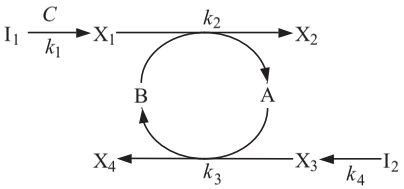
Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
3	Same assumptions as in model 1 except: I_1 and I_2 change linearly with time such that $I_1(t) = 80 + t$ $I_2(t) = 100 - t$	X_2 and X_4 are dynamically regulated by the levels of I_1 and I_2 as described in model 1. Switching of dependence on the inputs at a point beyond the similar point determined by the steady-state analysis is observed. This time lag is due to an accumulation of X_3. Concentrations of A(t) and B(t) show a step function with the same time lag as X_2 and X_4.	The dynamic behavior of the cyclic enzyme system display catastrophic behavior in response to specific changes in external input. The system can realize a neuronic model capable of storing memory.		77,82
4	Same assumptions as in model 1 except: I_1 and I_2 are represented by a sinusoidal function with time t, such that $I_1(t) = 10 + 2\sin\left(\frac{2\pi}{40}t + \frac{\pi}{2}\right)$ $I_2(t) = 10 + 2\sin\left(\frac{2\pi}{40}t - \frac{\pi}{2}\right)$	Concentrations of A(t) and B(t) follow the pattern described in models 1 and 2. The time lag is observed due to accumulation of X_1 and X_3. Switching does not occur until the accumulation of either substrate is canceled.	The accumulated substrate is equivalent to a "condenser" and is applicable to a kind of "memory storage."		81
5	 Same assumptions as in model 4 except: $k_5 \neq 0$ and $k_6 \neq 0$.	No time lag is observed, and the conversion from switched on to off (or from off to on) occurs rapidly according to the difference in amount between I_1 and I_2.	The behavior of the system is equivalent to that of an electronic switching circuit.		81

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

6	 Same assumptions as in model 3 except: The reactions $X_1 \rightarrow X_2$ and $X_3 \rightarrow X_4$ are reversible.	Increase in the values of the rate parameters, k_{-1} and k_{-2}, leads to a more gradual rise of A(t) and B(t), and the step function is not obtained. When the ratio of k_1/k_{-1} and k_2/k_{-2} is fixed at 1, increase in the rate parameters leads to a sharper rise of A(t) and B(t).		82
7	Same assumptions as in model 3 except: Reaction mechanisms are represented by ordered bi–bi enzymic kinetics. All reaction steps are assumed to be reversible.	The dynamic characteristics of A(t) and B(t) are qualitatively similar to those observed in model 3. The initial concentrations of the enzymes and cofactors affected the dynamic characteristics of the system significantly. The switch is obtained only when these concentrations are over a certain threshold value.	The system can be used as a switching controller when it is coupled to the reaction $S \rightarrow P$ and a cofactor A is essential to produce P. Thus, A can be controlled by I_1 and I_2 and the switch properties will be obtained for P(t).	82
8	Same assumptions as in model 1 except: Two sinusoidal inputs with a phase difference θ between them: $I_1(t) = 10 + 2\sin\left(\frac{2\pi}{40}t\right)$ $I_2(t) = 10 + 2\sin\left(\frac{2\pi}{40}t - \theta\right)$	The concentration profiles obtained for A(t) and B(t) show the switching observed in model 4, but the on/off times depend on θ. The frequency and amplitude of X_1 and X_3 depend on θ.	Focusing on the dynamics of $X_1(t)$ and $X_3(t)$, the system can play the role of a rectifier circuit. The amount of rectification depends on θ.	80

(*continued*)

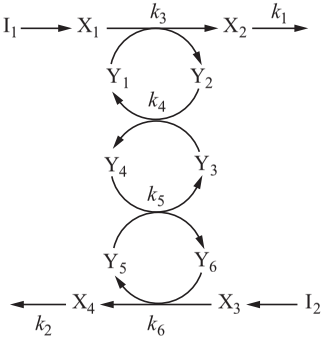
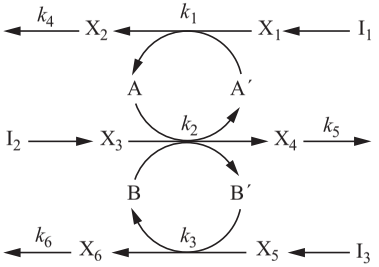
Table 1.1 (*continued*)

Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
9	Same assumptions as in model 4 except: Reaction mechanisms are represented by ordered bi–bi kinetics. All reaction steps assumed to be reversible.	As in model 4. Since the reaction involves several steps, the value of A(t) or B(t) when switched on was not the initial total concentration of them (1.0).			80
10	 I_1 and I_2 are represented by a sinusoidal function with time t. I_3 and I_4 are external inputs of A and B, respectively. k_5 and k_6 are degradation rate constants. $k_5 = k_6 = I_3(t) + I_4(t)$.	The switching time of the cyclic system can be regulated by I_3, I_4, as well as by I_1 and I_2. When a pulse of I_3 and I_4 is introduced, one can select the time of introduction in order for A or B to be switched-on thereafter. Introduction of I_3 or I_4 affect only the switching time of A and B, but not the oscillatory pattern of X_2 and X_4.		The comments concerning I_1 and I_2 are applicable to I_3 and I_4 as well.	83
11	 Concentrations of I_1, I_2, X_2, and X_4 are held constant. C is the input parameter. All the reactions are reversible.	A and B evolve in time to a unique steady state dictated by C. Steady-state concentrations of A and B show step functions in respect to C. k_2 and k_3 determine the steepness of the jump. When $k_2 \neq k_3$ the curves of the steady-state concentrations of A and B are not symmetric.	The system can act as a chemical neuron in which the concentration of A or B determines the state of the neuron (fire or quiescent).	The mathematical equations for this model also agree with the case of batch reactions. Here the step $I_1 \rightarrow X_1$ is a catalytic reaction, and the step $I_2 \rightarrow X_3$ is a noncatalytic one.	109, 116

[a]These observations are those of the present authors.

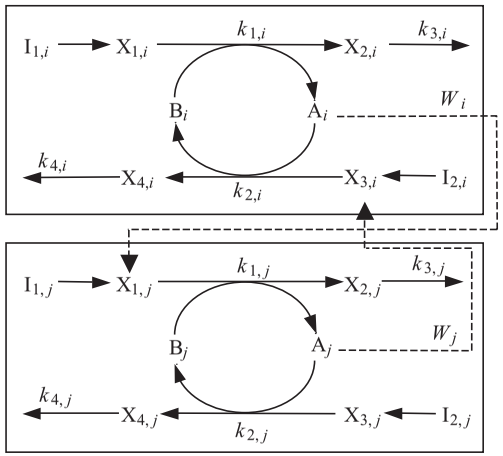
P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

Table 1.2 Models of Biochemical Networks Based on the Cyclic Enzyme System

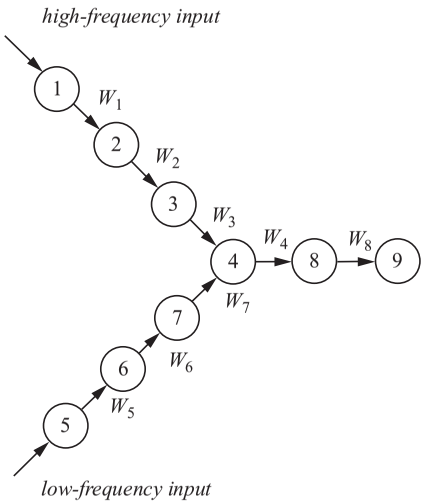
Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
1	 Coupled cyclic enzyme system: I_1 and I_2 are inputs to the system's pools of substrate X_1 and X_3, respectively; simple mass action kinetics; irreversible reactions.	A stepwise change in the steady-state concentration of $Y_i(t)$ is observed. The results are similar to those obtained using a monocyclic enzyme system (described in model 1 in Table 1.1).	The system can be applied for examination of control mechanisms of metabolic coupled enzyme systems, such as the sugar transport system in bacteria.	The mathematical equations for this model and for the following ones agree with the case of batch reactions, in which there is no mass flow into or out of the system. However, I_1 and I_2 cannot be defined as mass flows if volume changes are not considered.	79
2	 Bicyclic enzyme system: A, A′, B, and B′ are cofactors; I_1, I_2, and I_3 are constant inputs.	The steady-state concentrations of A and B ($\bar{A}$, $\bar{B}$) are determined by the minimum input among I_1, I_2 and I_3: I_1 minimum: $\bar{A}, \bar{B} = 0,1$ I_2 minimum: $\bar{A}, \bar{B} = 1,1$ I_3 minimum: $\bar{A}, \bar{B} = 1,0$ I_1 and I_3 minimum: $\bar{A}, \bar{B} = 0,0$	Based on the results presented, the basic logic functions NOT, AND, OR can be built using monocyclic or dicyclic enzyme systems.	The comments concerning I_1 and I_2 are applicable to I_3 as well.	81

(*continued*)

Table 1.2 (*continued*)

Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
3	 The basic element is similar to the one assumed in model 1 in Table 1.1. The *j*th element is assumed to have an excitatory or inhibitory affect on the *i*th element according to the following options: (a) Excitatory interactions: A_i affects $X_{1,j}$ $\frac{dX_{1,j}}{dt} = (I_{1,j} + W_i A_i) - k_{1,j} X_{1,j} B_j$ (b) Inhibitory interactions: A_j affects $X_{3,i}$ $\frac{dX_{3,j}}{dt} = (I_{2,i} + W_j A_j) - k_{2,i} X_{3,i} A_i$ (c) Reversible interactions: both excitatory and inhibitory.	The number of excited elements in sequentially connected systems is related proportionally to the values of the excitatory stimulus. When the introduction of the excitatory stimulus is too late, it can not be transmitted. The excitatory stimulus is amplified to a certain limit and attenuated during propagation. By assuming several excitatory stimuli and varying their frequencies, the long-term potentiation phenomenon can be observed. Supposing reversible interactions between two elements, a continuous switching pattern of the output is observed.	One can interconnect basic elements excitatorally, inhibitorally, or reversibly and construct large networks.	The species A_i and A_j play the role of effector for another enzymic reaction, and their concentrations are not affected by this activity.	84,85

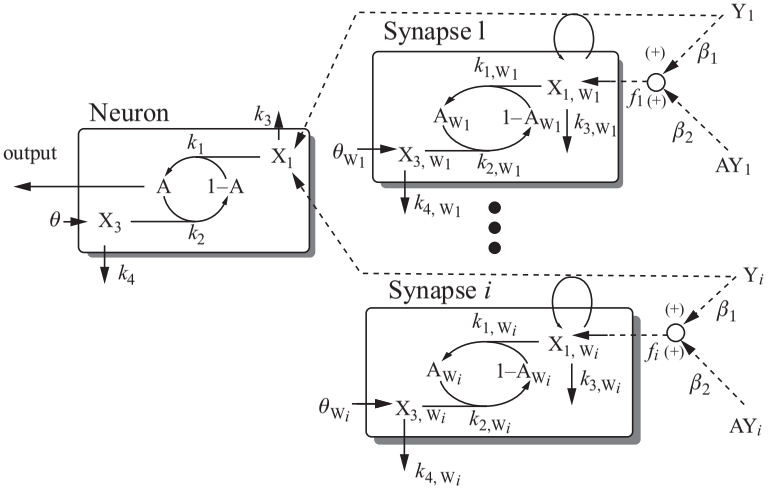
P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

4	External stimuli on a branched series of excitatory interactions, mentioned in model 3.  	High-frequency excitatory stimuli which were introduced to the first element was amplified and transmitted to the ninth element. Low-frequency excitatory stimuli which was introduced to the fifth element was attenuated during propagation leading to selective elimination of synaptic connection between the seventh and fourth elements.	The system shows the physiological phenomenon termed *selective elimination of synapses* generally produced as a result of a low-frequency train of electrical stimuli to the synapses.	85
5	External stimuli on a branched series of excitatory interactions, mentioned in models 3 and 4 except in the basic element the substrates $X_{1,i}$ and $X_{3,i}$ do not accumulate and are removed with k_5 and k_6 (model 5 in Table 1.1).	Selective elimination of synapses cannot be observed.	Neural network model composed of formal neurons without the capacity of memory storage cannot be applicable to the study of information processing of real neural networks.	85

(*continued*)

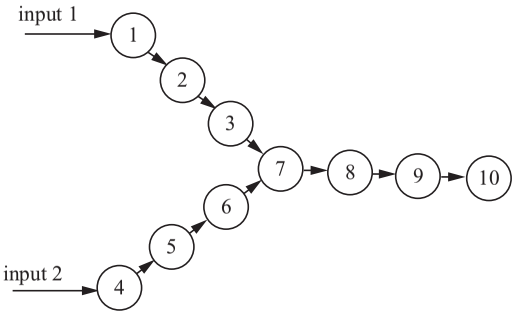
Table 1.2 (*continued*)

Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
6	excitatory input signal				86



$X_{1,Wi}$: synaptic efficacy for excitatory input Y_i at synapse i
A(t): the output signal
θ: threshold value
β_1, β_2: arbitrary coefficients
f_i: feedback factor from output A
$f_i = (\beta_1 + \beta_2 \mathrm{A})\mathrm{Y}_i; \quad i = 1, 2, \ldots, n$
$k_3, k_4 \gg k_{3,\mathrm{W}_i}, k_{4,\mathrm{W}_i}$

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

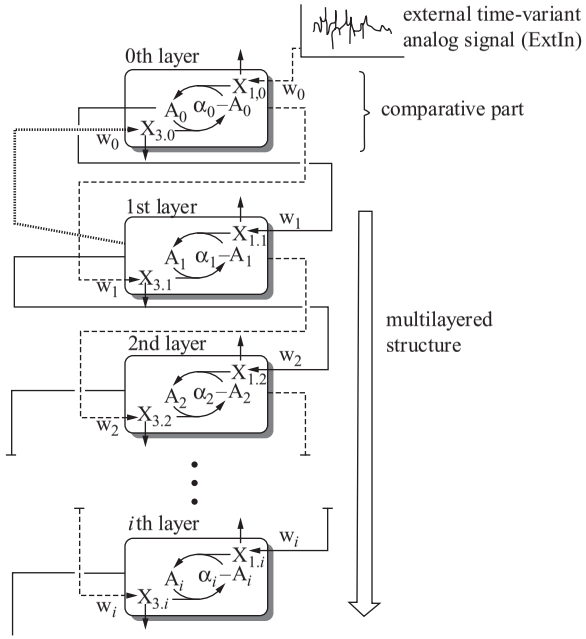
7	 Each element is assumed to be composed of the basic scheme described in model 6, and to transmit the signal $A_i(t)$ to the subsequent one. When $0 < t < 60$, high-frequency input is introduced to the first element and low-frequency input is introduced to the fourth. When $60 < t < 120$, low-frequency input is introduced to the first element and high-frequency input is introduced to the fourth.	Excitatory high-frequency stimuli which were introduced to the first element during $0 < t < 60$ are amplified and transmitted successfully to the tenth element. Excitatory low-frequency stimuli which were introduced to the fourth element during $0 < t < 60$ are attenuated during propagation leading to selective elimination of synaptic connection. During $60 < t < 120$, excitatory high-frequency stimuli which were introduced to the fourth element turned out favorably, leading to the revival of the signal path from the fourth to the seventh, and the low-frequency stimuli introduced to the first element caused selective elimination of synaptic connection between the third and the seventh elements.	High-frequency activation of excitatory synapses produces a long-lasting increase in synaptic efficacy, and excitatory stimuli with low frequency are unfavorable for the growth of synaptic efficiency.	86

(*continued*)

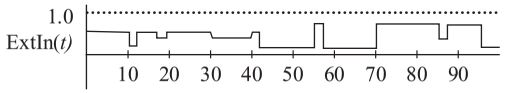
P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

Table 1.2 (*continued*)

Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
8	Assumptions 1 to 6 as in model 6. The number of synapses denoted by i is 2. X_{1,W_1}, X_{1,W_2}: synaptic efficacies of the test path and conditioning path, respectively. Low-frequency test input and no conditioning input.	The test input itself has not caused long-term potentiation of the synaptic efficacy (X_{1,W_1} or X_{1,W_2}).			86
9	Assumptions 1 to 3 as in model 8. Low-frequency test input and high-frequency conditioning input are positively correlated. After the in-phase inputs are introduced 14 times only, the test input is reintroduced and the changes in X_{1,W_1} and A were investigated.	The synaptic efficacy X_{1,W_1} is potentiated during a long time period.			86
10	Assumptions 1 to 3 as in model 9. Low-frequency test input and high-frequency conditioning input are anticorrelated.	The synaptic efficacy X_{1,W_1} is weakened or depressed, leading to long-term depression of synaptic strength.			86



α_i: threshold value for cutting off the signals. At the 0th layer, external analog signal ExtIn(t) and output of the first layer $A_1(t)$ are introduced to $X_{1,0}$ and $X_{3,0}$ respectively. Outputs of the $(i-1)$th layer, are the input of the ith layer. The external signal, ExtIn(t), is represented by a step function with time t:



$\alpha_i = 1$, $i = 0,1,2,\ldots,6$.

The time courses of ExtIn(t) and $A_1(t)$ are almost the same except for the oscillatory behavior of $A_1(t)$. $A_0(t)$ behaves as a control variable which changes in value so as to minimize the function J:

$$J = \int_{t_0}^{t_f} [\mathrm{ExtIn}(t) - A_1(t)]^2 \, dt$$

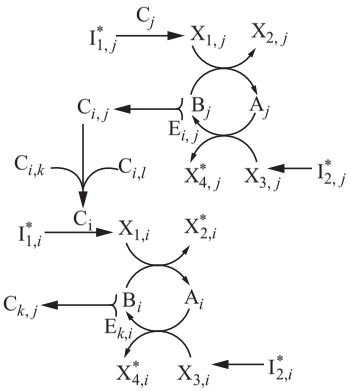
After the second layer the time courses of A_i (t) are filtrated by the threshold value $\alpha_i/2$ and gradually settled to a two-state, having the values 0 and 1.0 only, and having a higher signal-to-noise ratio.

The biochemical neuron has another role as a "transducer" of external analog signals to impulse signals, where the external signals are received at the receptive field and transduced to impulse signals by cutting off at a certain threshold value.

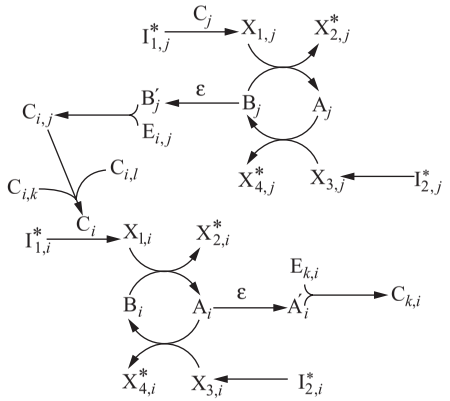
(*continued*)

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

Table 1.2 (*continued*)

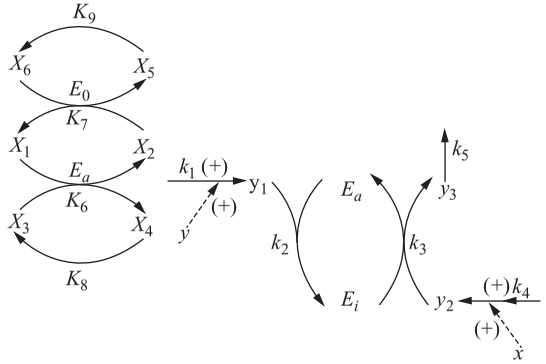
Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
12	Assumptions 1 to 3 as in model 11. The external analog signal, ExtIn(t), has a uniform random value between 0 and 1.  $\alpha_i = 1.8$, $i = 0,1,2,\ldots,5$, $\alpha_6 = 1.0$.	External random input signals are filtrated by the threshold value $\alpha_i/2 = 0.9$ and transformed into impulse signals.	By changing the α_i values, any time-variant external analog signal can be filtrated by an arbitrary threshold value.		86
13	 Each neuron is as in model 11 in Table 1.1. Neurons are chemically distinct. The effect of one neuron on the others is contained in C_i. Neuron i is affected by neurons j and k. A_j and B_j are activators of an enzyme $E_{i,j}$ to form $C_{i,j}$. The enzyme-activator reaction is fast and at equilibrium. A_j, A_k: inputs; C_i: output; $E_{i,j} \ll 1$ and $C_i = \sum_j C_{i,j}$.	For excitatory connections: $(E_{i,j} + A_j \rightleftharpoons C_{i,j})$: $C_{i,j} = \frac{E^0_{i,j}}{1+\frac{1}{kA_j}}$ For inhibitory connections: $(E_{i,j} + B_j \rightleftharpoons C_{i,j})$: $C_{i,j} = \frac{E^0_{i,j}}{1+\frac{1}{k(A_0-A_j)}}$ where k is the equilibrium constant. By adjusting the values of $E^0_{i,j}$ and k, neuron i can perform logic operations on the state of neurons j and k.	Various types of logic gates can be constructed when the threshold value for C_i is defined as 1: AND, OR, NOR, A_j AND NOT A_k.	Concentrations of A_i are set at $t = 0$ and the output is obtained at steady state. No time dependence is considered.	109

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

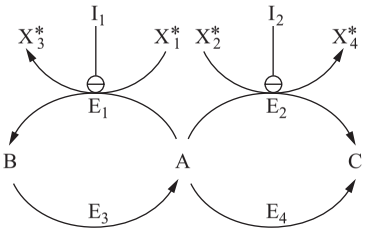
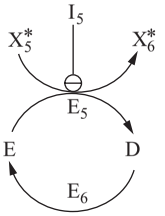
14	 Assumptions 1 to 5 as in model 13. The state of the chemical neuron is allowed to change only at discrete times, dictated by an autonomously oscillating catalyst ε. The concentration of ε is very small except during short intervals. ε interacts with A_j or B_j of each neuron, and rapid equilibrium occurs when its concentration is large. $C_{i,j}$ can be activated/inhibited by more than one species: $E_i + A'_j \leftrightharpoons C_i$ $E_i + A'_k \leftrightharpoons (E_i A'_k)$	By changing the number of neurons, the form of the connections between them, and the definitions the concentrations that represent inputs and outputs to the system, various finite-state machines can be specified.	A binary decoder, a binary adder, and a stack memory can be built.	110

[a]These observations are those of the present authors.

Table 1.3 Models of Other Biochemical Systems

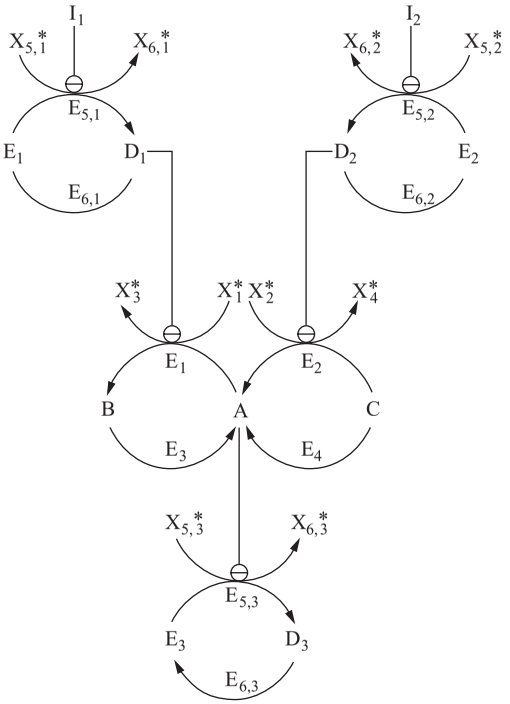
Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
1	 E_a: active enzyme E_i: inactive enzyme x,y: excitatory and inhibitory factors, respectively; both factors remain constant during the reaction E_0: an enzyme with constant activity All the reactions are first order. Inputs: x and y; outputs: x_1 and x_2.	The steady-state concentrations of E_a and E_i show step functions with respect to the value of x/y: $E_a(x, y) = [0; x \geq y, 1; x < y]$ $E_i(x, y) = [1; x \geq y, 0; x < y]$ The concentrations of x_1 and x_2 change in a similar way except for the appearance of a curved corner whose magnitude seems to depend on the concentration of E_0.	The enzymic conjugate system described can realize the two-factor model. The system was included as a control element in a feedback system. In this case, a specific configuration of the control element can maintain the value of the end product at a desired level.		76
2	$A^* \xrightarrow{E_1} B \longrightarrow P^*$ The concentrations of A and P are held constant. The conversion of B to P follows Michaelis–Menten kinetics. I_1 and I_2: two external effectors of E_1 Output: steady-state concentration of B Input: concentration of I_1 and I_2.	Three different mechanisms for the kinetics of E_1 can be used to construct three different logic gates: AND, OR, and XOR. The degree of cooperativity in the binding of E_1 and I_1 or I_2 determines the steepness of the transition from low to high steady-state concentrations of B.			115

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

3	 E_1 to E_4 are irreversible enzymes that follow Michaelis–Menten kinetics. E_1 and E_2 are inhibited by the noncompetitive inhibitors I_1 and I_2. Concentrations of X_i are held constant. Inputs: concentrations of I_1 and I_2. Output: steady-state concentration of A. The concentrations of the species marked with (*) are fixed.	When no inhibitors are present, the steady-state concentrations of A, B, and C are equivalent. When one of the inhibitors is present, the material is apportioned between A and one of the other species. When both inhibitors are present, conversion of A to the other species is blocked. The steepness of transition between the highest and lowest concentrations of A, the values of these concentrations, and the symmetry of the response depend on the kinetic parameters of the enzymes.	The system can function as a logical AND gate.	115
4	 E_5, and E_6 are irreversible enzymes that follow Michaelis–Menten kinetics. E_5 is inhibited by the noncompetitive inhibitor I_5. Concentrations of X_i are held constant. Inputs: concentrations of I_5. Output: steady-state concentration of D. The concentrations of the species marked with (*) are fixed.	The concentration of D is high (low) when the concentration of I_5 is low (high). The steepness of transition between the highest and lowest concentrations of D and the value of this concentration depend on the kinetic parameters of the enzymes.	The system can function as a logical NOT gate.	115

(*continued*)

Table 1.3 (*continued*)

Model No.	Model Characteristics	Results	Conclusions and Applications	Comments[a]	Refs.
5	 Three NOT gates (model 4), and one AND gate (model 3). Inputs: concentrations of I_1 and I_2. Output: steady-state concentration of D_3.	The output reaches its maximum value when one of the inputs or both of them are present in significant amounts. The output is minimized when neither input chemical is present.	The system can function as a logical OR gate.		115

[a]The observations are those of the present authors.

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

The works presented in Tables 1.1 to 1.3 [76–86,109–122] deal only with theoretical aspects of the enzymic biochemical devices, and the biochemical devices were not carried into practice. Moreover, Okamoto [85] suggests using silicon technology instead of biomaterials for practical implementation of the device based on the cyclic enzyme system.

This study is also based on the cyclic enzyme system, but its leading concept is to accomplish practical implementation of this system using biomaterials. In this respect, the analytical models developed here are related to several biochemical reactors in which enzymic reactions take place. This practical approach cannot be found in the models reviewed [76–86,109–122].

1.5 OSCILLATIONS IN BIOCHEMICAL SYSTEMS

Many oscillatory patterns can be found in biological systems [123–126]. It is generally recognized in engineering that encoding information in a frequency provides resistance to degradation by noise and enhanced precision of control. Rapp [124] suggested that many biological oscillations can be envisaged to reflect the biochemical implementation of this control strategy.

Intracellular communication often proceeds in a pulsatile, rhythmic manner [126]. Moreover, an increasing number of hormones are found to be secreted in a pulsatile manner, and the physiological efficiency of these signals appears to be closely related to their frequency [126]. Based on this understanding, a number of classes of drug therapies have been shown to require a periodic, pulsatile regimen of delivery for efficacy or optimization [131], and several delivery strategies have been proposed to respond to this need [127–131].

1.6 KINETIC CHARACTERISTICS OF CYCLIC ENZYME SYSTEMS

Many examples of enzymatic cyclic systems have been developed in practice [87–107]. These systems can be utilized to construct the biochemical device proposed by Okamoto et al. [76–86]. The kinetic properties of five enzymes that catalyze reactions in which cofactors are required, and therefore can participate in a cyclic enzyme system, are summarized in Table 1.4 [132–144]. These enzymes are glucose-6-phosphate dehydrogenase (G6PDH, E.C. 1.1.1.49), glutathione reductase (GR, E.C. 1.6.4.2), glucose dehydrogenase (GDH, E.C. 1.1.1.47), L-lactate dehydrogenase (LDH, E.C. 1.1.1.27), and alcohol dehydrogenase (ADH, E.C. 1.1.1.1).

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan

Table 1.4 Kinetic Properties of Enzymes Used in Cyclic Systems

Enzyme	Process	K_m values	Conditions	Reaction Mechanism	Other Findings	Refs.
G6PDH From brewer's yeast	Glucose-6-phosphate + NADP → gluconate-6-phosphate + NADPH	$K_{m,\text{G6P}} = 6.9 \times 10^{-5}$ M $K_{m,\text{NADP}} = 3.3 \times 10^{-5}$ M In the presence of $MgCl_2$ 0.01 M: $K_{m,\text{G6P}} = 5.8 \times 10^{-5}$ M $K_{m,\text{NADP}} = 2.0 \times 10^{-5}$ M	0.063 M Tris buffer, pH 8 at 25°C		The enzyme is inhibited by NADPH, which is competitive with NADP. The inhibition constant $K_I = 2.7 \times 10^{-5}$ M. The reaction is reversible and the equilibrium constant is $6 \pm 0.7 \times 10^{-7}$ M at 28°C.	132
From *Candida utilis*		$K_{m,\text{G6P}} = 2.3 \times 10^{-4}$ M $K_{m,\text{NADP}} = 6.7 \times 10^{-5}$ M	93 mM glycine–NaOH buffer pH 9.1, also containing 9.3 mM $MgCl_2$ and 0.93 mM EDTA			133
GR From baker's yeast	Oxidized glutathione + NADPH → reduced glutathione + NADP	$K_{m,\text{GSSG}} = 6.1 \times 10^{-5}$ M $K_{m,\text{NADPH}} = 7.6 \times 10^{-6}$ M	Phosphate buffer, pH 7.6 at 25°C			134
From sea urchin egg		$K_{m,\text{GSSG}} = 1 \times 10^{-4}$ M $K_{m,\text{NADPH}} = 5 \times 10^{-6}$ M	0.1 M potassium–phosphate buffer, pH 7.2, containing 1 mM EDTA		Addition of 1 mM EDTA increases the enzyme activity. Further addition of EDTA shows no further effect.	135

GDH From beef liver	Glucose + NAD → glucono-δ-lactone + NADH	$K_{m,\mathrm{NAD}} = 4.3 \times 10^{-6}$ M pH | $K_{m,\mathrm{glucose}}$ (M) 6.28 | 34.9×10^{-2} 7.00 | 3.13×10^{-2} 8.92 | 32.6×10^{-2}	0.05 M phosphate buffer, pH 7.6 at 21–22°C		The reaction is reversible and the equilibrium constant is $2.9–3.3 \times 10^{-7}$ M.	136
From ox liver		$K_{m,\mathrm{glucose}} = 15 \times 10^{-2}$ M $K_{m,\mathrm{NAD}} = 1.5 \times 10^{-5}$ M	0.05 M phosphate buffer, pH 7		The reaction is reversible and the equilibrium constant is 30×10^{-7} M at pH 7.	137
		$K_{m,\mathrm{glucose}} = 7 \times 10^{-2}$ M	0.05 M phosphate buffer, pH 7.6			
From rat liver		$K_{m,\mathrm{glucose}} = 0.3–0.7$ M $K_{m,\mathrm{NAD}} = 0.38\ \mu$M $K_{m,\mathrm{NADP}} = 0.45\ \mu$M	Phosphate buffer, pH 8.2			138
From *Bacillus Megaterium*		$K_{m,\mathrm{glucose}} = 47.5 \times 10^{-3}$ M $K_{m,\mathrm{NAD}} = 4.5 \times 10^{-3}$ M $K_{i,\mathrm{NAD}} = 69 \times 10^{-5}$ M	Acetate–borate buffer, pH 9 at 25°C	Ordered Bi–Bi		139
LDH From rabbit muscle	Pyruvate + NADH → lactate + NAD	$K_{m,\mathrm{pyruvate}} = 5.2 \times 10^{-5}$ M				140

(*continued*)

Table 1.4 Kinetic Properties of Enzymes Used in Cyclic Systems (*continued*)

Enzyme	Process	K_m values	Conditions	Reaction Mechanism	Other Findings	Refs.
LDH From rabbit muscle	Pyruvate + NADH → Lactate + NAD	$K_{m,\text{pyruvate}} = 15 \times 10^{-6}$ M $K_{m,\text{NADH}} = 35 \times 10^{-7}$ M $K_{\text{PN}} = 6.5 \times 10^{-12}$ M^2	0.05 M sodium phosphate buffer, pH 6.8 at 25°C	Ordered Bi–Bi. $\frac{V_m}{V} = 1 + \frac{K_{m,\text{P}}}{[\text{P}]} + \frac{K_{m,\text{N}}}{[\text{NADH}]} + \frac{K_{\text{PN}}}{[\text{P}][\text{NADH}]}$	The reaction is reversible and the equilibrium constant is 2.76×10^{-12} M at pH 7 and 25°C. Pyruvate is an inhibitor.	141
		$K_{m,\text{pyruvate}} = 1.64 \times 10^{-4}$ M $K_{m,\text{NADH}} = 1.07 \times 10^{-5}$ M $K_{\text{PN}} = 1.38 \times 10^{-9}$ M^2	0.25 M phosphate buffer, pH 6.8 at 25°C		Pyruvate and lactate inhibit the enzyme with $K_{\text{I,pyruvate}} = 2.02 \times 10^{-4}$ M $K_{\text{I,lactate}} = 0.209$ M	142
From *Lactobacillus plantarum*	Pyruvate + NADH → lactate + NAD	$K_{m,\text{pyruvate}} = 3.7 \times 10^{-4}$ M	0.1 M Tris buffer, pH 8			143
ADH From baker's yeast	Ethanol + NAD → acetaldehyde + NADH	pH $K_{m,\text{NAD}}$ (mM) $K_{m,\text{Eth.}}$ (mM) $K_{i,\text{NAD}}$ (mM) 4.9 0.224 107 0.390	0.01 M acetic acid–sodium acetate buffer, pH 4.9	Ordered Bi–Bi	The reaction is reversible and the equilibrium constant is 0.98×10^{-11} M at 25°C	144
		5.95 0.106 43 0.340 7.05 0.108 26 0.270 8.1 0.118 18.5 0.385	0.1 M phosphate buffer, pH 5.95, 7.05, 8.1			
		8.9 0.150 10 0.860 9.9 0.200 5 2.40	0.01 M glycine-NaOH buffer, pH 8.9, 9.9			

P1: OTA/XYZ P2: ABC
c01 JWBS019-Filo November 4, 2009 7:29 Printer Name: Sheridan