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Simulative Representation of Biological Knowledge using Object-oriented Database Language

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Abstract

Major advances in bio-technology enable us to describe various phenomena occuring in the body using the language of genes and proteins. It is important to represent these phenomena in knowledge base, and to visualize them properly. The visualization of the phenomena with reference to related databases facilitates researche on genes.

As the first step in realizing a database like the one stated above, we have studied the representation of biological knowledge needed to describe biological phenomena and have developed a prototype knowledge base. The knowledge base is described in micro-Quixote, an object-oriented database language executable on Unix. The knowledge base can cover the knowledge related to signal transduction within a cell and that related to transcription of genes.

In our prototype system, a sort of simulation can be done. With the arrival of a signaling ligand at the surface of a cell, proteins along suitable pathways are activated in our simulated cell. As a consequence of series of activations(a chain of inferences), some biological responses are deduced and shown to users.

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1 Introduction

Amazing advances in bio-technology now allow us to describe a range of phenomena that occurs in the human body by applying the language of genes or DNA. Genes encode the proteins that constitute the body. Proteins act not only as the building blocks of the body, but also serve to regulate it. These proteins are also coded in the genes. Consequently, a knowledge of genes and proteins is essential to understanding phenomena such as the immune system.

There is now enough information describing biological phenomena so that we can explain biological phenomena to some extent. To do this, however, we must collect information from several sources, biological text books [Alberts 1994], papers and databases[Bairoch 1992]. These sources differ in their authors' interests and in the way they abstract information. Recently, several researchers [Goto 1993] have attempted to integrate biological databases. Their works are a necessary step toward the description of biological phenomena. However, all have been interested mainly in the integration of data and have paid little attention to the representation of biological phenomena.

As a result, non-experts in biology find it difficult to integrate these information sources to adequately grasp biological phenomena. Sometimes, even experienced biologists fail to get an integrated view of biological phenomena, because many biologists are only able to keep up with progress in their specialty.

It is important to express, in a knowledge base, the phenomena played by genes and proteins and to visualize these phenomena adequately. The use of visualization helps students of biology to understand biological phenomena in the body. Also, visualizing phenomena by referring to related databases facilitates research on genes and gives biologists further inspiration for new research.

As a first step toward a knowledge base like that described above, we have studied representations of biological knowledge needed to describe biological phenomena, especially signal transduction pathways, and have developed a prototype knowledge base and display system. From our experience with object-oriented knowledge bases[Hirosawa 1993; Tanaka 1993], we thought that an object-oriented knowledge base would be suitable for describing biological concepts. We decided to employ micro-Quixote, an object-oriented database language [Yokota *et al.* 1993], in this project. We expected, through the use of the object-oriented language, that concepts such as cell could be described naturally. In related research area, [Karp 1994] studied representations of metabolic pathways using Lisp.

In this paper, we describe knowldge representation in our prototype knowledge base system. In the next section, we describe the processes occuring in the body. Both intracellular and intercellular processes are explained. Then, our prototype knowledge base, which represents the processes inside as well as outside a cell, is presented. Finally, an example of showing how the knowledge base can be used is presented.

2 What happens in the body

Because the body is composed of cells, if cells and the interaction between cells can be described properly, it should be possible to describe the biological phenomena in the body. This section explains the intracellular and intercellular processes that occur in the body.

2.1 Intercellular process

Cells in the body communicate with each other. They secrete hormones such as insulin and growth factors such as EGF. When cells receive some chemical substance, be it a hormone or growth factor, they may or not exhibit some reaction, depending on the type of the cells. When some reaction occurs, some intracelluar processes take place.

Fig. 1 is a simple example of an intercellular process. The figure shows the intercellular process between an insulin secretory cell (C1), such as a beta cell of the pancreas, and an insulin target cell (C2), such as a muscle cell.

An insulin secretory cell secretes insulin(Reaction R1) when it receives glucose(Stimulus to cell S1). An insulin target cell takes in glucose (Reaction R2) when it receives insulin (Stimulus named S2). When a hormone such as insulin is secreted, it spreads thoroughout the body. So, the insulin secreted by an insulin secretory cell travels around the body to arrive at an insulin target cell. Then, the insulin target cell receives insulin to take in the glucose around it. To sum up, stimulus (reception of glucose) to an insulin secretory cell (S1) results in some reaction (intake of glucose) by the insulin target cell (R2).

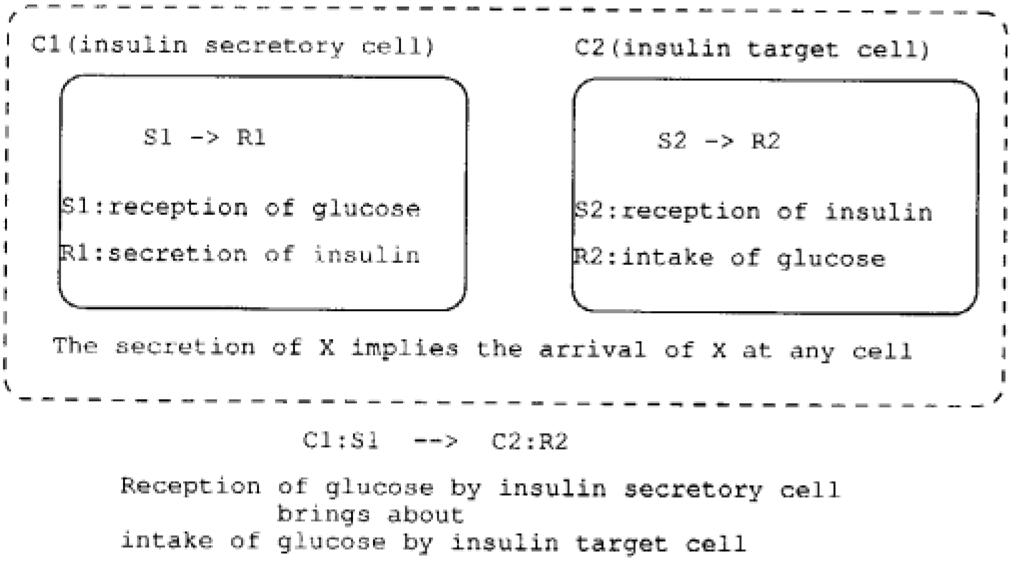

C1(insulin secretory cell)
C2(insulin target cell)
S1 -> R1
S1:reception of glucose
R1:secretion of insulin
S2 -> R2
S2:reception of insulin
R2:intake of glucose
The secretion of X implies the arrival of X at any cell
C1:S1 --> C2:R2
Reception of glucose by insulin secretory cell
brings about
intake of glucose by insulin target cell

Fig. 1

2.2 Intracellular process

As mentioned in the previous subsection, a cellular process can be simply described as a stimulus-reaction pair ($S1$(reception of glucose)$- >$ $R1$(secretion of insulin) in C1). However, many intracellular processes exist between S1 and R1. Biologists often want to determine and understand these intracellular processes.

Fig. 2 is an example of an intracellular process. To describe intracellular processes, two kinds of knowledge, transcription knowledge and intracellular process knowledge, are necessary. We will explain the significance of these two kinds of knowledge later.

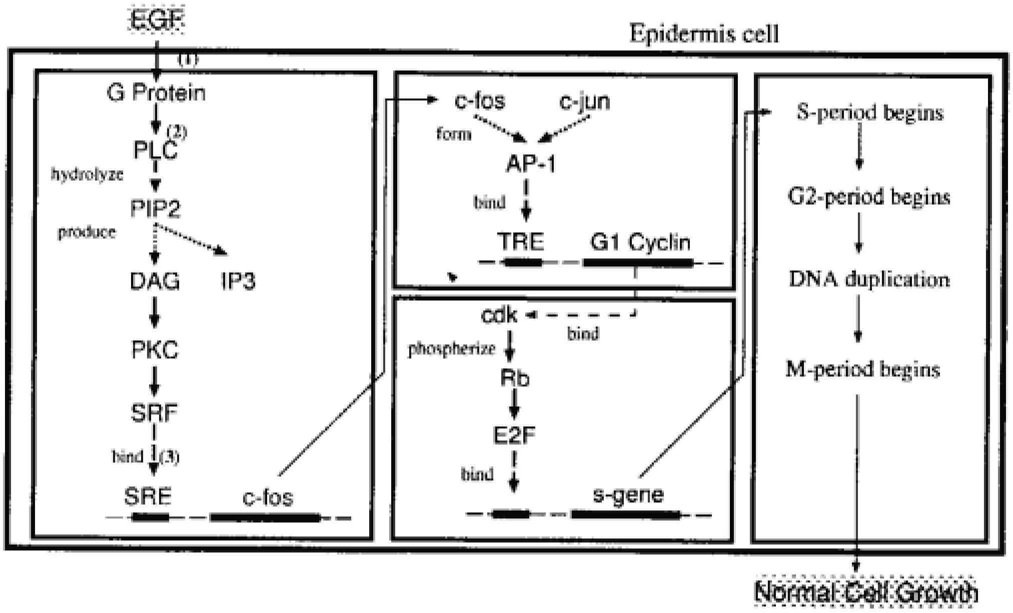


Fig. 2

In this case, the stimulus to the cell is the reception of EGF. The corresponding reaction by the cell is normal cell growth. EGF(Epidermis Growth Factor) belongs to the growth factor family. When a growth factor is received by some types of cell, the cell is duplicated as the result of intracellular processes. In the case of EGF, the epidermis cell duplicates upon receiving EGF.

In the figure, a solid arrow from one entity to another indicates the activation of the latter by the former. For example, the reception of EGF activates G protein (1), then the activation of G protein activates PLC (2). A dashed arrow signifies the process indicated beside the arrow. For example, the activation of SRF enables SRF to bind to SRE(3).

When EGF arrives at the epidermis, after a series of processes, SRF binds to SRE. Then, the c-fos gene, controlled by SRE, is expressed and c-fos is produced. The produced c-fos and c-jun form AP-1 to bind to TRE. Then, the G1 Cyclin gene, controlled by TRE, is expressed and G1 Cyclin is produced (SRE and TRE are referred to as response elements). After a series of processes, the s-gene is produced. After S-period, DNA duplicatation occurs in G2-period. Finally, normal cell growth occurs after the M-period.

The knowledge necessary to describe an intracellular process like the above can be classified into two classes. One class relates to which response element controls which gene (SRE controls c-fos, TRE controls G1 Cyclin, and RE(s-gene) controls s-gene). We refer to such knowledge as "transcription knowledge". The other class, named “intracellular process knowledge”, is knowledge other than transcription knowledge. For example, knowledge (1) (reception of EGF activates G protein), knowledge(2) and knowledge(3) are examples of intracellular process knowledge.

3 Prototype knowledge base

In this section, we explain the prototype knowledge base. An overview of the system is shown in Fig.3. It is supported by any Unix machine.

The system is divided into two modules, the knowledge base module and the display module. The result of inference performed by the knowledge base module is transferred to the display module and the result, like that shown in Fig.2, will be displayed using the GUI. The display module is coded in C, is executable on X11R5, and is programmed based on Motif.

The knowledge base module is written in the object oriented database language, micro-Quixote [Yokota *et al.* 1993] executable on UNIX. It is composed of an inference module and five knowledge bases, namely the intracellular process knowledge base, transcription knowledge base, cellular process knowledge base, intercellular process knowledge base and cell inheritance knowledge base. The inference module utilizes the deductive feature of micro-Quixote.

These knowledge bases are explained below.

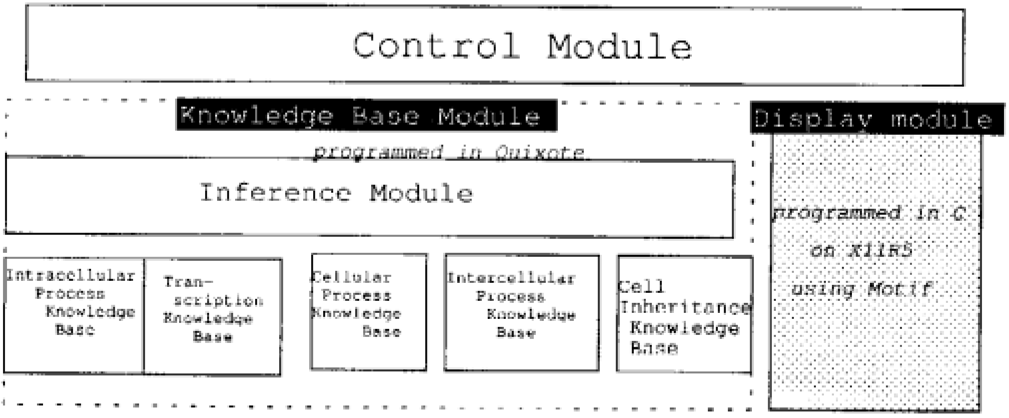

Control Module
Knowledge Base Module
programmed in Quixote
Inference Module
Intracellular Process Knowledge Base
Tran-scription Knowledge Base
Cellular Process Knowledge Base
Intercellular Process Knowledge Base
Cell Inheritance Knowledge Base
Display module
programmed in C on X11R5 using Motif

Fig.3

Intracellular Process Knowledge Base

The following is a portion of the intracellular process knowledge base. Processes within a cell can be described using knowledge in the intracellular process knowledge base and transcription knowledge base. Each entry in the intracellular knowledge base represents individual processes inside a cell. For the sake of simplicity, only simple knowledge is shown.

```
receive[name="EGF"]/[result=increase[name="DG"]];;   (1)
increase[name="DG"]/[result=active[name="PKC"]];;    (2)
active[name="PKC"]/[result=active[name="SRF"]];;     (3)
```

"EGF", "DG", "PKC" and "SRF" are proteins or a sort of protein. The knowledge can be read as follows. If "EGF" is received, "DG" is increased (1). If "DG" is increased, "PKC" becomes active (2). If "PKC" is active, "SRF" becomes active (3). The description is done in the form "A/[result = B];;". It signifies that if A is satisfied, then B becomes true. In this way, individual processes within a cell can be described.

In the three knowledges (1),(2) and (3), 'receive', 'increase' and 'active' are objects in micro-Quixote. However, we could choose "EGF","DG","PKC" and "SRF", which correspond to entities in the entity-relationship model. Database researchers might regard the latter as being a more suitable choice. However, when we describe intracellular processes, it is essential that we describe possible events. We selected 'receive', 'increase' and 'active' as objects because possible events can be described using these words.

To understand the collective result of each process, let us assert knowledge " receive[name="EGF"]" to the knowledge base. If we ask whether "active[name="SRF"]" is true, the knowledge base answers "yes". To prove "active[name="SRF"]" the above three knowledges are used. An important point that must be noted here is that only individual processes are described in the knowledge base and that the proof of "active[name="SRF"]" is the result of a series of inferences. Here, the deductive feature of micro-Quixote is used.

Transcription Knowledge Base

The entries in the transcription knowledge base describe which response element controls what gene. Two examples are shown below. The knowledge can be read as follows. SRE controls c-fos (4) and TRE controls G1 Cyclin (5).

```
gene[name="SRE",coded="c-fos"];;     (4)
gene[name="TRE",coded="G1cyclin"];; (5)
```

Cellular Process Knowledge Base

Processes within a cell can be described using knowledge in the intracellular process knowledge base and transcription knowledge base. The reaction of a cell to a specified stimulus can be described theoretically using the two knowledge bases. However, it is often the case that some intracellular processes, necessary to deduce some reaction to a specified stimulus, have not yet been discovered. In such cases, knowledge that directly relates stimulus to reaction is neccesary. The cellular process knowledge base describes the stimulus-reaction relationship.

Two examples are shown below. Knowledge can be read as follows. When IL-4 and IL-5 arrive at a B cell, the B cell receives IL-4 and IL-5 (6). When a B cell receives IL-4 and IL-5, it secretes IgE (7). *If it did not have knowledge (6), a B cell would not be able to secrete IgE, even when IL-4 and IL-5 arrive at the B cell.*

```
arrive[name1="IL4",name2="IL5",cell="B cell"]/
  [ result= receive[name1="IL4",name2="IL5",cell="B cell"]];; (6)
receive[name1="IL4",name2="IL5",cell="B cell"]
   /[result=secrete[name="IgE",cell="B cell"] ];; (7)
```

Intercellular Process Knowledge Base

The intercellular process knowledge base describes the interaction between cells. There are many types of interaction. However, the prototype knowldege base has only one entry for the intercellular process knowledge base. The knowledge (8) shown below is very important and it can be applied to a large portion of intercellular processes. This means that chemical substance P, secreted by some cell named C1, can arrive at any cell named C2.

```
secrete[name=P, cell=C1]/
     [ result= arrive[name=P,cell=C2]];; (8)
```

Cell Inheritance Knowledge Base

The cell inheritance knowledge base describes the hierarchical relationship between classes of cells. By means of the knowledge base, the intracellular process knowledge and/or intercellular process knowledge described in some class of cell is inherited by its lower class. In micro-Quixote, inheritance of knowledge between classes can be neatly realized by the use of the module concept.

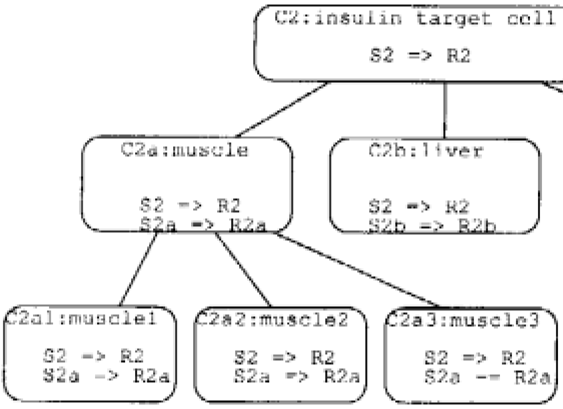
Two examples are shown in Fig.4. Knowledge (9) means that liver, muscle and fat cells are subclasses of the insulin_target_cell. Muscle has muscle1, 2 and 3 as its subclasses (knowledge (10)). In the language of micro-Quixote, liver, muscle and fat cells are sub-modules of the insulin_target_cell. In this case, class and module have almost the same implication.

In this case, if intracellular/intercellular process knowledge represented as $S2 => R2$ in Fig.4, is assigned as information in the insulin_target_cell using the module concept, the information is inherited by its three subclasses. As a result we don't have to describe the relationships in the three subclasses thanks to the inheritance of the object-oriented database language.

```
insulin_target_cell >- {liver,muscle,fat_cell};; (9)
muscle >- {muscle1,muscle2,muscle3};;            (10)
```

In micro-Quixote, for example knowldge(a) ..... knowledge(n) are assigned as knowledge belonging to the module named liver, if we use the following notation:

liver::{ knowledge(a) ...... knowledge(n) }



S => R means knowledge neccssary to describe processes between stimulus S to reaction R.

Fig.4

4 Example of execution

In this section, we give an example of execution of our knowledge base. In this case, "what happens if EGF arrives at an epidermis cell" is asked (a0). The knowledge base answers that normal cell growth occurs (a1). The knowledge base also indicates which intracellular processes occur between the reception of EGF (a2), and normal cell growth (a28). The intracellular process is almost the same as that explained as an example of an intracellular process using Fig.2.

Among a series of processes(a2 ~ a28), there are three processes corresponding to the gene expression for producing protein(a11,a15,a19). The three processes are expression of c-fos, G1 cyclin and s-gene. The three processes are derived from the transcription knowledge base. Other processes are derived from the intracellular process.

After three gene expressions, the cell enters the s-period (a20) then the G2-period (a23). After cell duplication (a24), the cell enters the G2-period (a27) to complete normal cell growth (a28).

```
?- arrive[name="EGF",cell=epidermis]/[result=Result,path=Path].         (a0)
?- yes.
Result = normal_cell_growth,                                            (a1)
Path = state[now= receive[name="EGF",cell=epidermis],                   (a2)

  next=state[now= active[name="G protein",cell=epidermis],              (a3)
  next=state[now= active[name="PLC",cell=epidermis],                    (a4)
  next=state[now= hydrolysis[name="PIP2",cell=epidermis],               (a5)
  next=state[now= produce[name=list[element1="DAG",element2="IP3"],
                                                   cell=epidermis],(a6)
  next=state[now= increase[name="DAG",cell=epidermis],                  (a7)
  next=state[now= active[name="PKC",cell=epidermis],                    (a8)
  next=state[now= active[name="SRF",cell=epidermis],                    (a9)
  next=state[now= bind_element[name="SRE",cell=epidermis],              (a10)
  next=state[now= express[name="c-fos",cell=epidermis],                 (a11)

  next=state[now= form[name="c-jun",with="c-fos",cell=epidermis],       (a12)
  next=state[now= produce[name="AP-1",cell=epidermis],                  (a13)
  next=state[now= bind_element[name="TRE",cell=epidermis],              (a14)
  next=state[now= express[name="G1cyclin",cell=epidermis],              (a15)

  next=state[now= active[name=cdk,with="G1cyclin",cell=epidermis],      (a16)
  next=state[now= phosphorize[name="Rb",cell=epidermis],                (a17)
  next=state[now= active[name="E2F",cell=epidermis],                    (a18)
  next=state[now= expresse[name="s-gene",cell=epidermis],               (a19)

  next=state[now= begin[name="s-period",cell=epidermis],                (a20)

  next=state[now= phosphorize[name="Cdc2 kinase",cell=epidermis],       (a21)
next=state[now= bind[name="Cdc2 kinase",to=cyclinB,cell=epidermis], (a22)

next=state[now= begin[name="G2-period",cell=epidermis],              (a23)
next=state[now= duplicate[cell=epidermis,object="DNA"],,             (a24)
next=state[now= active[name="Cdc25 phosphatase",cell=epidermis],,    (a25)
next=state[now= active[name="Cdc2 kinase",cell=epidermis],,          (a26)

next=state[now= begin[name="M-period",cell=epidermis],,              (a27)

next=normal_cell_growth]]]]]]]]]]]]]]]]]]]]]]]]]]                    (a28)
```

5 Discussion

1. So far, little attention has been paid to the representation and visualization of biological phenomena. In our system, possible events within a cell(e.g. If "EGF" is received, "DG" is increased) are stored in the knowledge base. If we want to know the events that occur if some stimulus comes from outside the cell, and if we ask so, inducable events are successively calculated in the system. The series of events that occurs after the stimulus is then shown to the user. This can be regarded as being a simulation of the phenomena that occur in the body. Through the simulation, users can experience the biological processes happening in the body. Also, they can understand which proteins play a role in the phenomena.

   An important point that must be noted is that only individual processes are described in the knowledge base and that the result of normal cell growth is derived as a result of a series of inferences. Here, the deductive feature of micro-Quixote is used.

2. In this knowledge base, we utilize inheritance of the object-oriented database language to effectively describe the reaction of a cell to a stimulus received from outside. Specifically, the module concept of micro-Quixote is used. Because information on the processes that occurred between the stimulus and the reaction to the stimulus described for any class of cell is automatically inherited by its lower classes, there is no need to describe the information in those lower classes. This reduces the amount of knowledge necessary to describe biological phenomena.

3. We can describe the reaction of a cell to a stimulus from outside in two ways. In one way, we can describe it by describing every intracellular process involved in producing the reaction to the stimulus. Alternatively, we can describe it by describing only the stimulus-reaction relationship. We can therefore see biological phenomena in different levels of abstraction.

   Often, biologists want to understand biological phenomena in greater detail. This, for example, happens when they are interested in intracellular processes. It also happens when they want to compare two biological processes and try to gain information through analogy. In this case, the former way of description is preferable.

However, it happens that not every intracellular process necessary to describe the reaction to a stimulus hasn't identified. In this case, biological phenomena can be described only in the latter way. And this type of description is also useful when users are interested in intercellular interactions

4. To make the system more powerful, we believe that knowledge to explain protein, e.g. EGF, is neccessay. We are now investigating how to construct such a knowledge base and how to connect our system to existing databases.

5. In this paper, the example of simulation result is output as language because we focused on representation of biological knowledge with an object-oriented database language. With the display module in Fig.3 [Hirosawa *et.al forthcoming*] the system can visualize the simulation result. With the graphic interface, users can view simulation results in a few levels of abstraction.

   Visualizing the simulation result will enhance our understanding of biological phenomena in the body, with the comparison of similar processes giving biologists a deeper insight into biologial phenomena. We believe that our system will contribute to the progress of biology and medicine.

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