REPLACEMENTS FOR ANIMAL EXPERIMENTATION, COMPUTER SIMULATION MODELS

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ABSTRACT

For demonstration of animal experiments of medical and paramedical faculties, a large number of animals are sacrificed every year. Such demonstrations can be humanely presented using computer simulations rather than experimental animals. For example, software 'X-coley: An Interactive CD-ROM for Pharmacology Undergraduates' was developed. It has become increasingly difficult to perform animal experiments, because of issues related to the procurement of animals, and strict regulations and ethical issues related to their use. A number of computer simulation models (CSMs) are available. Interactive demonstrations were conducted to encourage the departmental faculty to use CSMs. CSMs can be used repeatedly and independently by students, and this avoids unnecessary experimentation and also causing pain and trauma to animals. Recently a hybrid model based on the finite element method and on a compartmental biophysical representation of peripheral nerve fibers and intraneural electrodes was developed. Animal experiments were performed using the same stimulation protocol as in the computer simulations in order to rigorously validate the model predictions and understand its limitations. This software displays complete video demonstrations of different procedures like isolation and mounting of animal tissues followed by on screen interactive interface to study the effects of various drugs on the isolated tissues. It simulates / demonstrates experiments causing undue pain to animals. The presentation is made user friendly and self explanatory.

Keywords: Alternative of animal, simulation model, computer science.
INTRODUCTION

It is difficult to perform animal experiments, because of issues related to the procurement of animals, and strict regulations and ethical issues related to their use. Computer modeling can replace certain kinds of animal use, particularly in education. High school biology classes, for example, might practice dissection on a computer model rather than on real, live frogs. Even medical schools are beginning to develop "virtual reality" devices for students to practice on. You can find an example of a "Virtual Frog Dissection Kit." Plastic models and realistic manikins also can take the place of live animals for some educational purposes.¹¹

Software is great help in understanding concepts of pharmacology and experimentation research by students. These can significantly reduce the number of animals used for demonstration and research work.

Computer simulations are useful in evolutionary biology for hypothesis testing, to verify analytical methods, to analyze interactions among evolutionary processes, and to estimate evolutionary parameters. Plenty of computer simulators have been developed to simulate DNA sequence data with recombination.⁷

Computers are also limited by their processing power. A recent simulation of just half a mouse’s brain required the use of the world’s fastest supercomputer – Blue Gene/L (300% faster than the second fastest super computer). The vast complexity of the simulation meant that it was only run for 10 seconds at a speed ten times slower than real life – the equivalent of one second in a real mouse brain. The researchers say that although the simulation shared some similarities with a mouse’s mental make-up in terms of nerves and connections it lacked the structures seen in real mice brains.

Multimedia CD-ROMs, and models are also more economical than traditional animal-based teaching exercises. Whereas the “traditional” approach involves the acquisition and disposal of animals on an ongoing basis, purchasing a set of CD-ROMs represents a one-time expenditure for a product that can be used repeatedly for many years. Schools can save tens of thousands of dollars each year by implementing reusable replacements for animal “specimens”. Computational techniques used to model, analyze and simulate signalling networks. Role of signalling networks in three major areas: signal transduction, cellular rhythms and cell-to-cell communication Aims: To provide opportunities for students to learn, develop and practice:⁶
• The application of the scientific method and experimental design.

• Problem-solving skills.

• Generic biological practical skills (e.g. weighing, preparing solutions, pipetting).

• Skills in observation, measurement, appreciation of variability, concern for precision, data handling skills: analysis, presentation and interpretation.

• Oral and written communication skills: report writing, poster production.

• An appreciation of safety in the laboratory.

• Pharmacology-specific practical skills.

• Concepts of pharmacology – re-enforcing existing and gaining new knowledge.

These computer models allow managers to better understand the spatial and temporal visitation patterns and “experiment” with different management strategies. Specifically, simulation models can be used to:

• Improve communication of implications of management prescriptions to the public

• Better understand the baseline spatial and temporal patterns of visitor use.

• Predict how distributions of visitor use are likely to change in response to both management actions and factors not subject to managerial control.

• Monitor hard-to-measure parameters (such as people at one time at a certain attraction or walking on particular trails) by using easily measured indicators (such as number of cars entering the area or parking at a trailhead).

• Support the planning and management of visitor use in situations where monitoring and predicting visitor flow are difficult.

• Test the feasibility and effectiveness of management plan alternatives.

**Computer simulation**

Examples of computer simulations available include models of asthma, though potential new medicines identified using these techniques are currently still required to be verified in animal and human tests before licensing. Computer operated mannequins, also known as crash test dummies, complete with internal sensors and video, have replaced live animal trauma testing for automobile crash testing. The psychomotor skills that a student acquires in a training course in experimental pharmacology may not be directly relevant to the future tasks that a practising doctor has to undertake.

Computer models have been constructed to model human metabolism, to study plaque build-up and cardiovascular risk, and to evaluate toxicity of drugs, tasks for which animals are also
used. In 2007, US researchers using the world's fastest computer at the time, BlueGene L, modeled half a mouse's brain for just 10 seconds. However, due to limitations in computing power, the simulation could only be run at 1/10th the speed of an actual mouse brain.\textsuperscript{11,13}

Computer simulations do actually yield acceptable models in research on diabetes, asthma, and drug absorption, although potential new medicines identified using these techniques must still be verified in animal and human tests before licensing.

Mahatma Gandhi said, ‘We should not inflict cruelty on even the meanest of creatures. I also will have to answer for this in the court of the Almighty.’ No single person ever in the history contributed so much wealth to protection of animals as Ms. Hildegard Doerenkamp from Germany, the founder of Doerenkamp-Zbinden Foundation. The way of funds and expertise high-quality research that advances the 3Rs for development of pedagogic tools computer modeling for teaching, drug development, basic biomedical research, product testing, etc.\textsuperscript{3}

The following facilities are being built:\textsuperscript{3}

- A repository of dissection CDs, videos, mannequins, models, etc.
- A state of the art computational lab.
- in vitro testing facility and a cell line bank.
- Bioinformatics tools such as predictive toxicology (Q)SAR, Read Across, ebTrack, etc., for in silico analysis

Computer simulation achieved the same level of knowledge as those working in the laboratory and that primary learning objectives were equally well achieved by both approaches.\textsuperscript{2} Computer simulations, which are now widely available at relatively low cost, can provide a dry lab experience that may fulfil some, but not all, of the objectives of the animal labs.\textsuperscript{15}

The Computer Laboratory Demonstrations

A computer assisted learning (CAL) program was used specifically to offer an alternative, student centred approach to teaching the module. The CAL program is an easy-to-use laboratory simulation software and lab manual that consists of exercises containing a number of physiology lab activities that can be used to supplement or substitute wet labs. The CAL program allows us to repeat labs as often as we like, perform experiments without harming live animals, and conduct experiments that are difficult to perform in a wet lab environment.
because of time, cost, or safety concerns. Pre-lab and Post-lab Quizzes for each activity and Stop & Think and Predict Questions within the steps of each experiment help students make the connection between the activities and the physiological concepts they demonstrate. The CAL group also had a 2.5 hour introductory lecture. The lecture also included a brief introduction to describing the computer simulation.\textsuperscript{15}

Dynamic System

\begin{itemize}
  \item Molecular dynamic
  \item biochemical network
  \item population dynamics
  \item Gene-regulatory Networks
  \item signal networking
  \item metabolic networks
  \item Signal Transduction
  \item Cellular rhythms
  \item Intracellular signaling
  \item Receptor kinase electro
  \item Signaling cascade physiology circadian cell cycle healing system rhythms regulation
\end{itemize}

\textbf{Figure 1: An ontological view of biological dynamic systems to which modelling approaches have been applied.}\textsuperscript{1}
Modeling approaches and software
Simulation modeling software is needed to process input variables, generate data analyses, and produce output. Three approaches to modeling and simulation of relevance to recreation travel simulations are trace, probabilistic, and rule-based agent models. The same software program used for development of the IO model may be used to simulate clinical trial response data. The key element is the ability to add random (stochastic) variability to the model as residual error (measurement and model misspecification error) and IO model parameter variability. More realistic simulations will also include stochastic variation in sampling subject specific values from the covariate distribution model, and in the deviations from the nominal protocol defined by the trial execution model. Standard statistical software. e.g. SAS and S-Plus (MathSoft, Seattle, WA), or more specialized programs such as NONMEM and Pharsight Trial Designer—can accommodate all of these features. General statistical software is least convenient because all components of the simulation model must be created by the user. NONMEM has a comprehensive library of pharmacokinetic models but requires the user to create a data file to specify doses, covariates, and observation times for each subject.

Current aspects of computer simulation

- Computer simulations do actually yield acceptable models in research on diabetes, asthma, and drug absorption, although potential new medicines identified using these techniques must still be verified in animal and human tests before licensing.
- Other non-animal simulators have been developed for military use to mimic battlefield-induced traumas or to simulate hemorrhaging, fractures, amputations and burns. But all these processes are very many orders of magnitude simpler than the most simplified version of a nucleus in the brain.
- We produce a mathematical model and algorithmically describe how the model evolves over time. In other words, we write a computer program that runs a mathematical model of a process.
- We can do simulations that are astonishingly accurate. For example, we can do wind tunnel simulations of aircraft designs, and the results of those simulated experiments are perfect to within our ability to measure them.
- It can reduce the number of animals that are used in medical experimentation. But it can never replace them.
Table no. 1 survey about existing alternative methods in biomedical education\textsuperscript{2,4, 9, 10}

<table>
<thead>
<tr>
<th>Alternative</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Anatomy/dissection</strong></td>
<td></td>
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<tr>
<td>Comparative anatomy</td>
<td>- a collection of nine interactive learning modules covering the organ systems and related structures of mammals, birds and fish</td>
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<tr>
<td></td>
<td>- each module contains gross, histological and electron microscopic images</td>
</tr>
<tr>
<td>The rat – a functional anatomy</td>
<td>- understanding of anatomy and physiology in mammals from gross morphology to microscopic detail</td>
</tr>
<tr>
<td></td>
<td>- position of mammals and other classes within the vertebrate kingdom</td>
</tr>
<tr>
<td>The rat – a practical dissection guide</td>
<td>- a highly professional twenty minute full colour video of a practical dissection of rat</td>
</tr>
<tr>
<td>Vertebrate dissection guides – the pigeon</td>
<td>- examines the live pigeon, the external features and initial dissection techniques</td>
</tr>
<tr>
<td></td>
<td>- provides a detailed investigation of the external and internal features</td>
</tr>
<tr>
<td>Vertebrate dissection guides – the dogfish</td>
<td>- examines the live dogfish, the external features and initial dissection techniques</td>
</tr>
<tr>
<td></td>
<td>- provides a detailed investigation of the external and internal features</td>
</tr>
<tr>
<td><strong>Anesthesia</strong></td>
<td></td>
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<tr>
<td>Anesthesia of rats</td>
<td>- teaches students the basics and advanced skills of anesthetics</td>
</tr>
<tr>
<td></td>
<td>- simulates the complex technique of anesthetics</td>
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<tr>
<td>Remote</td>
<td>- trains the skills needed for patient monitoring during several (micro) surgical techniques</td>
</tr>
<tr>
<td>Virtual anesthesia machine</td>
<td>- web-based simulation of the flow of oxygen, nitrous oxide, carbon dioxide and volatile</td>
</tr>
<tr>
<td><strong>Laboratory animal science</strong></td>
<td></td>
</tr>
<tr>
<td>Animal experiment</td>
<td>provide research personnel with background information on animal experiments and alternatives</td>
</tr>
<tr>
<td></td>
<td>- supports researchers to carefully plan an experiment, including taking the steps of thinking of alternatives</td>
</tr>
<tr>
<td>Pictures instead of animals</td>
<td>provides moving pictures of laboratory animals displaying typical symptoms after being injected with pharmacological substances</td>
</tr>
<tr>
<td><strong>Pharmacology/toxicology</strong></td>
<td></td>
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<tr>
<td>Autonomic pharmacology</td>
<td>simulates experiments on the human eye to teach the effects of autonomic drugs (useful replacement for the Finkleman preparation)</td>
</tr>
<tr>
<td>Inflammation pharmacology</td>
<td>simulates a range of experiments designed to demonstrate the action of inflammatory mediators and pharmacological agents on the \textit{in vivo} inflammatory response in the anesthetized rabbit</td>
</tr>
<tr>
<td>Module</td>
<td>Description</td>
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<tr>
<td>Intestinal absorption</td>
<td>simulates experiments designed to demonstrate by investigation the important characteristics of the transport of two important nutrients – hexoses and amino acids, in the small intestine</td>
</tr>
<tr>
<td>Langendorff heart</td>
<td>highly interactive and simulates experiments which may be performed on the isolated perfused mammalian heart (Langendorff preparation)</td>
</tr>
<tr>
<td>Respiration pharmacology</td>
<td>based on pulmonary function data obtained from guinea pig to teach the fundamental pharmacology of the airways</td>
</tr>
<tr>
<td>Neuromuscular pharmacology</td>
<td>simulates experiments performed on the sciatic nerve-anterior tibialis muscle preparation of the cat (in vivo) to illustrate the important differences in the pharmacological action of depolarizing and non-depolarizing blocking agents</td>
</tr>
<tr>
<td>Simulations of pharmacological experiments on the guinea pig ileum</td>
<td>covers basic receptor theory, including occupancy and how response size is related to occupancy - user is introduced to dose-response curves through simulated experiments of the action of drugs on isolated guinea pig ileum</td>
</tr>
<tr>
<td>Physiology/biochemistry/molecular studies</td>
<td></td>
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<tr>
<td>Blood coagulation</td>
<td>teach the physiology and laboratory assessment of haemostatic function and the use of the haemostatic profile in diagnosis of common clinical disorders</td>
</tr>
<tr>
<td>Muscle physiology</td>
<td>simulates experiments on the frog sciatic nerve – gastrocnemius muscle preparation to illustrate physiological properties of skeletal muscle</td>
</tr>
<tr>
<td>Frog heart</td>
<td>simulates a number of experiments which can be performed on the in situ heart of a pithed frog</td>
</tr>
<tr>
<td>Nerve physiology</td>
<td>simulates a number of experiments performed on the frog sciatic nerve preparation to illustrate some of the important properties of mixed nerves</td>
</tr>
<tr>
<td>Renal function in humans</td>
<td>- simulates experiments to study the renal function in humans</td>
</tr>
<tr>
<td>SimMuscle</td>
<td>simulates the classic experiment on the leg muscular of a frog (musc. gastrocnemius)</td>
</tr>
<tr>
<td>Biochemical simulations (enzyme assay, urea synthesis, peptide sequence)</td>
<td>contains seven simulated programs: enzyme assay, urea synthesis, peptide sequence, oxygen electrode, radio-immunoassay, nitrogen balance and energy balance</td>
</tr>
<tr>
<td>Cellular Respiration</td>
<td>program designed to teach, by investigation, the important principles of cellular respiration and is based on a series of experiments which may be performed on isolated rat liver mitochondria in vitro</td>
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Experiments in human neuro-muscular physiology introduces the student to some of the apparatus (oscilloscope, stimulator) and the experimental techniques used to examine resting potential, action potential and nerve conduction.

Veterinary physiology can be used as a teaching material in the lecture.

Squid axon simulates a number of experiments on the isolated giant axon of the squid, which may be performed to investigate the biophysical properties of nerve.

SimNerv simulates the classic experiment on the sciatic nerve of a frog (nerve stimulation and recordings of compound action potentials (CAP)) - offers a completely equipped virtual laboratory in a realistically looking 3D framework on the computer screen.

**Importance**

A wide range of use particularly as alternatives to animal labs in pharmacology, physiology, biochemistry, anatomy (dissection, vivisection, technical skills)\(^{15}\)

- precise results
- unlimited number of repetitions
- learning can take place independent of tutor support
- offer the ability to plan/design experiments (e.g. drug doses/combinations in pharmacology and physiology)
- high technical possibilities (broadcasting, video-projector presentations, virtual conferences)
- Simple Easy & Express
  
  For example, Simple Express can pick 10 people for drug testing and 2 people for alcohol testing in a single draw in one request.

- RandomWare Basic
  
  Employers and TPAs who need to print random selection notification letters, monitor year to date annual selection percentages, track positive and negative results and generate DOT MIS reports should consider Random Ware Basic.

- Random Ware Plus
  
  Resources for EDI data management (import raw Lab/MRO test result data), billing utilities, DOT MIS reports and tools to manage a consortium of clients for random selection will find solutions in Random Ware Plus. The Plus System will appeal to TPAs, MROs and
Occupational Health professionals as well as Employers with large employee populations across multiple locations.

CONCLUSION

It has become increasingly difficult to perform animal experiments, because of issues related to the procurement of animals, and strict regulations and ethical issues related to their use. The pre-clinical tests of the drug on the animals cause a large number of mortality. Due to this computer stimulation is an important tool for study of the drug. Computing science, is the ability to perform sophisticated model analysis which can better relate predicted behavior and observations.

REFERENCES

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