AUCTORES
Globalize your Research

International Journal of Clinical Case Reports and Reviews
Soroush Sardari *

Open Access

Review Article

Non-animal and Non-human Cancer Models for Drug Screening

Reza Hamed Rahimi [1,2], Soroush Sardari [1]*, Soheila Yaghmaei [2]

[1] Drug Design and Bioinformatics Unit, Department of Medical Biotechnology, Biotechnology Research Center, Pasteur Institute of Iran, Tehran, Iran.

[2] Group of Biotechnology, Department of Chemical and Petroleum Engineering, Sharif University of Technology, Tehran, Iran.

***Corresponding Author:** Soroush Sardari, Department of Medical Biotechnology, Pasteur Institute of Iran.

Received Date: July 03, 2023 | **Accepted Date:** July 14, 2023 | **Published Date:** July 21, 2023

Citation: Reza H. Rahimi, Sardari S., Yaghmaei S., (2023), Non-animal and Non-human Cancer Models for Drug Screening, *International Journal of Clinical Case Reports and Reviews*, 14(2ss); **DOI:10.31579/2690-4861/322**

Abstract:

Cancer remains a major source of morbidity and mortality despite decades of scientific and clinical research and trials of promising new treatments. It is one of the leading causes of death worldwide, estimated to be the cause of the deaths of more than 600,000 individuals in the United States alone. Even many years after the discovery of cancer and the numerous research undertaken on it, the identification of anticancer medications continues to be a difficult undertaking. In addition, the development of drug and multidrug resistance hinders drug development. Therefore, continuous drug screening and testing should be undertaken in order to discover the treatment for this disease. Consequently, extensive investigations into cancer models are required for drug screening. One of these models comprises silico models which are inaccurate, unclear, and not time-effective. Next is animal models, which cannot accurately anticipate human reactions and are expensive, time-consuming, and challenging to work with. Human models are alternative models that, despite their ability to accurately predict human behavior, are far more expensive, demanding, and unethical. However, there is yet another model known as the microbial model. They are less costly, less time-intensive, more manageable, simple to cultivate, and straightforward to work with. In this study, we look into the major flaws of animal and human models and provide a new and more effective method for testing anticancer medications and combating anticancer drug resistance.

Key words: cancer; cancer models; anticancer drug screening; microbial cancer models; drug resistance

Introduction

Cancer

Despite decades of scientific and clinical study and trials of promising new therapies, cancer remains a leading cause of mortality and morbidity. The expected number of cancer-related fatalities in the United States is around 600,000, while the number of new cases is approximately 1.9 million. This indicates that a person diagnosed with cancer has a 31.5 percent chance of passing away [1]. This ratio is so high that it makes cancer one of the world's deadliest diseases. Cancer is caused by a succession of changes in genes that alter the functionality of cells. Cancer disrupts cellular interactions and causes the malfunctioning of essential genes. This disruption alters the cell cycle and results in aberrant proliferation. Some of the primary sources of these alterations include exposure to chemical compounds, smoking, ambient chemical agents having carcinogenic qualities, and microorganisms such as viruses and bacteria [2]. Moreover, research [3] suggests that high-frequency electromagnetic radiation may also be linked to lymphatic and hematopoietic malignancies. Therefore, it is vital to design cancer-treatment medications.

Drug

It now costs $3 to $5 billion and 12 to 15 years to bring a single medicine to market [15], making drug development a tremendously costly endeavor. There are several cancer treatment methods. These techniques include chemotherapy, radiation treatment, hormone therapy, and targeted therapy, among others. These approaches are medical treatments that either directly target the death of cancer cells or the destruction of tumor tissue, or halt the multiplication of cancer cells [4]. Nevertheless, despite the discovery of cancer and the countless studies undertaken on it, it is still difficult to identify anticancer drugs, even after many years. In addition, tumors are capable of developing drug resistance, which makes therapy even more difficult. As technology develops and our understanding of cancer grows, scientists are able to create new, stronger treatments.

However, these treatments cannot be commercialized immediately since they must be studied and approved to determine the most effective treatment for each form of cancer. Before entering human clinical trials, lead candidate medicines generally undergo ADMET (absorption,

distribution, metabolism, excretion, and toxicity) evaluation in vitro and in vivo (in animals) [15]. Evaluation of the effectiveness and safety of novel medications and treatment techniques in a preclinical setting is essential to the drug development process. It is based on a wide set of in vitro, ex vivo, in silico, and in vivo experiments that are designed to anticipate the physiological responses of pharmacological treatments in people and consequently select the first implementation of a therapy [6].

Models

Silico Models

The relatively young field of Quantitative Systems Pharmacology (QSP) blends systems biology techniques with quantitative pharmacology methodologies. Combining computational and experimental methodologies with QSP approaches enables systems-level knowledge of the mechanism of action of medications while utilizing the gathered data on authorized and unsuccessful drugs. By merging computational and experimental methodologies, Quantitative Systems Toxicology (QST), a new paradigm for toxicity assessment, seeks to comprehend the detrimental effects of medications, from molecular modifications to phenotypic findings. QST strategies have been beneficial for improving dosage and dosing schedules, thereby potentially reducing the cost of Phase I and II clinical studies. A greater knowledge of biological reactions to medications can minimize ambiguities in species extrapolations and permit the prediction of treatment responses, taking into account the genetic diversity of the patient or the presence of preexisting disorders. The field of computational toxicology [17] aims to anticipate the probable detrimental effects of a drug based on its chemical structure [16].

Despite having certain advantages, such as being more cost-effective, enabling the discovery of multitarget drugs, and producing predictions that can be translated, these models also have several drawbacks [6]. The intricacy of molecular dynamics contributes to one of the limits of these approaches. Analysis timeframes for this approach range from hundreds of nanoseconds to microseconds, depending on the size of the simulated systems. The trouble with this is that the time duration, which can range from milliseconds to seconds, is frequently too short to evaluate protein folding. Consequently, this can result in "inadequate sampling" of protein conformations [18]. Ensuring that proper scoring functions and algorithms are employed could otherwise jeopardize molecular screening [18]. They ensure the integrity of the intestinal mucosa, nervous system, and blood flow, as well as the production of enzymes and transporters. However, they need advanced surgical techniques and equipment, making them inappropriate for some laboratories. [19]In direct in-situ research, absorption is evaluated by the disappearance of the medication from the gastrointestinal tract. Nonetheless, many investigations employ indirect assessments, evaluating intestinal absorption based on the pace at which medications appear in plasma, their excretion in urine, or the rate of commencement or degree of pharmacologic effect [19]. In the majority of cases, they are based on a fairly limited training set, ranging from a few compounds to around 30. This would imply that unless the training sets are highly diverse, the predictive nature of the models may be constrained [20] unless the training sets are very different.

Animal Models

Historically, animal models have been incapable of predicting human responses to medications and disease. The dynamic and diverse microenvironments of live animals have made in vivo experiments a regulatory necessity for validating preliminary experimental findings [5,6]. Accordingly, animal models are poor at generating predictions, but they can provide information about the safety and efficacy of medications that cannot be obtained from individual animal trials [5]. Animal experiments have more significant faults, making them a less desirable candidate for anticancer drug screening. One is the difficulty with research [6]. Some animal models are only applicable to initial tumors and not to later phases of tumor development [7]. Animal models are a poor option since the results of various tests vary, animals' physiology differs from that of humans, and their reproductive organ malignancies (cervical, ovarian, uterine, vaginal, and vulvar) are not comparable to those of humans [6,8].

Social and personal factors are an additional concern about animal models. The growth in society's interest in and engagement in animal ethics has resulted in greater oversight of animal research throughout the years [6]. There are several rules and laws that must be followed carefully, including the "three Rs." Russell and Burch published the three Rs, which stand for Replacement, Reduction, and Refinement of Animal Studies, for the first time in 1959. The elimination of (non-human) animal use in scientific research is referred to as Replacement. Reduction entails utilizing fewer animals by using better statistical methods and literature research, and Refinement means lowering animal suffering and enhancing their wellbeing [9]. The cost component of using animal models presents a third obstacle. Costs associated with animal model research include the authorized permission fee, acquiring animals from the breeder, lodging in animal facilities and enhanced holdings, painkillers, analgesics, and sterilized surgical supplies [6].

In vitro Human Models

For the preliminary screening of possible drugs, in vitro models entail the use of various cultures, such as cell cultures, tissue cultures, and organ cultures. These procedures are an essential alternative to animal testing [6] because they are simple, less costly, and require less time. To overcome the drawbacks of human and animal models, in vitro tissue models have been created to facilitate the systematic, repeatable, and quantitative study of pharmaceuticals. By removing or minimizing the requirement for earlier models, these ones can become platforms for more strictly regulated, high-throughput drug screening and for studies of pharmaceuticals [11]. Tissue culture is a helpful technique for studying clinically relevant problems, particularly those linked to illnesses, screening, and cell toxicity processes. In the case of pathologically generated tissue, it has an intriguing use in the assessment of therapeutic compounds that might potentially cure the malfunction [12]. In addition, 3D cell culture platforms are excellent for investigating the effectiveness and tolerability of different tissues in a physiological context. These platforms are simple to operate, do not require external pumps or valves, and may be used again [13]. Certain factors, however, must be considered in order to achieve stable in vitro function. In primary culture, these characteristics are largely associated with increased demands on tissue for proper survival and differentiation under in vitro conditions. Other things that are needed are the use of special substrates, growth agents, and soluble media supplements, some of which have complicated ingredients [12].

Current animal and 2-D cell culture models used in metastasis research and medication development are inadequate surrogates for human cancer physiology [12]. Indeed, in vitro systems have significantly improved our understanding of toxicological pathways. However, there have been published critiques of the prospects for the complete substitution of animal research with in vitro methods. To completely transfer human in vitro models, considerable time is necessary. Expert

panels could not yet provide a time frame for more sophisticated systemic in vivo testing, such as repeated-dose toxicity, carcinogenicity, and reproductive toxicity. There are also difficulties in including xenobiotic metabolism in in vitro assays, capturing interactions between cell types, extrapolating from in vivo doses to in vitro concentrations, simulating the effects of long-term exposures in vitro, and extrapolating from perturbed pathways or biomarkers in vitro to adverse effects in vivo [22].

Human Models

In addition to political and security concerns, research on any medication in humans is prohibited in the majority of states due to significant ethical constraints. Before a human trial can begin, all proposed human trials must also undergo a meticulous risk-benefit analysis and be approved by human ethical committees governed by tight regulations. In general, pharmaceutical corporations only undertake this costly study when there is a strong probability of profit. Unless safety and risk-benefit criteria are satisfied, there is little desire to do human research, especially in light of the negative political constraints associated with an unlawful status [10]. Therefore, it is absolutely impossible to conduct human trials to evaluate novel anticancer drugs.

In their purest form, clinical trials are meant to monitor the outcomes of human volunteers under "experimental" circumstances under the scientist's supervision. This differs from noninterventional study designs, such as cohort and case-control studies, in which the researcher only evaluates the exposure of interest without altering it. The four stages of a clinical trial, which are sometimes called "phases," are meant to test a drug's safety and maximum tolerated dose (MTD), as well as its pharmacokinetics, pharmacodynamics, and drug-drug interactions in humans [21].

Phase I (also known as dosage escalation or human pharmacology) is the initial study of a novel investigational drug in humans. Typically, open-label studies are conducted with a limited number of "healthy" and/or "diseased" participants. Eventually, the MTD, or the medication dose prior to the onset of dose-limiting toxicity, may be identified via a variety of statistical techniques. Phase II trials, often known as "therapeutic exploratory" trials, are typically bigger than phase I studies and involve a greater number of patients with the target condition. In addition to testing safety, pharmacokinetics, and pharmacodynamics, they may also be made to answer important questions for planning phase III trials, such as what the best doses, frequency of doses, methods of administration, and results are. The phase III trial, also known as a "therapeutic confirmatory," "comparative efficacy," or "pivotal trial," is conducted on a larger and frequently more diverse group of individuals to demonstrate or confirm that the treatment is effective and to determine and estimate the frequency of common side effects. Phase IV trials, also known as "therapeutic use" or "post-marketing" studies, are observational studies conducted on FDA-approved drugs to: 1) identify less common adverse reactions, and 2) evaluate cost and/or drug efficacy in diseases, populations, or doses comparable to or significantly different from the original study population [21].

Microbial Models

Animals and other in vitro models are being replaced by microbiological models, which have been widely used in drug screening in recent years. For instance, the mouse model is one of the most often used models for researching microbial diseases. However, utilizing rats as infection models has ethical, financial, and logistical challenges. Firstly, keeping a sufficient number of animals to collect statistically significant data is costly and frequently considered unethical. Second, mammalian reproductive periods are protracted, which slows the progress of experiments. As an alternate model for studying microbial illnesses, Galleria mellonella has been introduced. G. mellonella larvae are readily available, affordable, and simple to employ as they require no specialized laboratory equipment. Their lack of ethical restrictions and short lifespan make them ideal for large-scale research [23].

The objective of anticancer drug development is to identify chemicals that selectively kill or limit the growth of tumor cells while leaving normal cells unaffected. Establishing the molecular distinctions between tumor cells and normal cells aids in the attainment of this selectivity. Consequently, the ultimate goal in cancer research [14] is to harness these genetic distinctions to produce novel anticancer medicines. Accordingly, microbial models, and especially fungal models, may be viable choices for anticancer drug screening. Saccharomyces cerevisiae, for instance, is one of the most basic eukaryotic creatures. It has a 90-minute life cycle, is cheap to maintain and cultivate, and is stable in both haploid and diploid forms. Its haploid genome is short and very simple, consisting of sixteen well-characterized chromosomes. Due to these characteristics, the yeast genome was the first eukaryotic genome to be sequenced, and its 6466 open reading frames are readily usable. Yeast has become an important model for human illnesses and biological processes. At least 31% of the yeast-encoded proteins have human orthologs, and almost 50% of the human disease genes have yeast orthologs. Consequently, the budding yeast Saccharomyces cerevisiae is frequently utilized as a model for studying fundamental processes that are applicable to all living creatures. Many of these systems, including cell cycle progression, DNA replication and segregation, preservation of genomic integrity, and stress responses, are altered by genetic and epigenetic modifications in cancer. Thus, yeast emerges as an appealing model for anticancer drug research [14].

	Model	*Advantages*	*Disadvantages*	*Time and Money*[1]	*References*
Silico Models	Protein based models	Cost-effective, Multitarget drug discovery, Translatable predictions	Intricacy of molecular dynamics, Too short time durations for protein folding evaluation, Possibility of jeopardizing molecular screening, Needing advanced surgical techniques and equipment, Limited training sets	5	6, 16-20

Animal Models (in vivo)	Mice, rabbits, pigs, etc.	Dynamic and diverse microenvironments, Provide information of safety and efficacy of medications	Difficulty with research, Social and personal factors, Not cost-effective	2	5-9
Human Models (in vitro)	2D and 3D cell and tissue cultures	Simple to operate, Less costly, Require less time, Repeatable, Investigating the effective ness and tolerability of tissue	Increased demand on tissue for different conditions, Complicated growth agents, Complicated soluble media supplements, Considerable time for complete transformation from animal models, Difficulties in including xenobiotic metabolism, Capturing interactions between cell types, Extrapolating from *in vivo* doses to *in vitro* concentrations, Simulating the effects of long-term exposures *in vitro*, Extrapolating from perturbed pathways or biomarkers *in vitro* to adverse effects *in vivo*	3	6, 11-13, 22
Human Models (Clinical Trials)	Phase I, Phase II	Good for testing drug's safety and MTD, Good for pharmacokinetics, pharmacodynamics, and drug-drug interactions in humans	Political concerns, Security concerns, Ethical constraints, Very costly	1	10, 21
Microbial Models	Microorganisms (fungi)	No ethical limitations, Cost-effective, No logistical challenges, Faster experimentations, Affordable, Simple to employ, No specialized laboratory equipment, Short lifespan which makes them ideal for large-scale research, Short and simple genomics, Appealing model for cancerous cells	Not ideal for testing drug safety and MTD	4	14, 23

Table 1: Description of various models used in drug studies among scientific community.

[1]The scoring system is based on 1 to 5. The higher the value the more feasible and cost effective the method is.

Conclusion

Due to the growth of drug and multidrug resistance in cancer and other diseases, different and diverse medications and medication regimens must be evaluated, necessitating a large number of test participants in order to identify the most effective therapy. Among the options and models that we have, we should choose the most optimal in terms of cost effectiveness, time for preparing the model, being easy to work with, having high similarity to cancer cells, and having a similar microenvironment to that of cancers. Therefore, by considering the advantages and limitations of each model, in order to address the growth of drug and multidrug resistance in the treatment of cancer and other diseases, microbiological models are ideal for evaluating alternative medications and pharmaceutical techniques.

References

1. American Cancer Society. (2022). Cancer Facts & Figures, *American Cancer Society*. www.cancer.org.
2. Hassanpour S H, & Dehghani M. (2017). Review of cancer from perspective of molecular. *Journal of Cancer Research and Practice.* 4(4): 127–129.
3. Ozdemir F, & Kargi A. (2011). Electromagnetic Waves and Human Health. *Electromagnetic Waves*.
4. Wang J-J, Lei K-F, & Han F. (2018). Tumor microenvironment: recent advances in various cancer treatments. *European Review for Medical and Pharmacological Sciences.* 22(12): 3855–3864.
5. Greek R, & Menache A. (2013). Systematic Reviews of Animal Models: Methodology versus Epistemology. *International Journal of Medical Sciences*. 10(3): 206–221.
6. Fontana F, Figueiredo P, Martins J P, & Santos H A. (2020). Requirements for Animal Experiments: Problems and Challenges. *Small*. 17(15): 2004182.
7. Gengenbacher N, Singhal M, & Augustin H G. (2017). Preclinical mouse solid tumour models: status quo, challenges and perspectives. *Nature Reviews Cancer*. 17(12): 751–765.
8. Basic Information About Gynecologic Cancers | CDC. (2021). www.cdc.gov.
9. Ritskes-Hoitinga M, & van Luijk J. (2019). How Can Systematic Reviews Teach Us More about the Implementation of the 3Rs and Animal Welfare? *Animals*. 9(12): 1163.
10. Christie M. (2012). Human testing of illicit drugs – the highs and lows. *The Conversation*.
11. Elliott N T, & Yuan F. (2011). A Review of Three-Dimensional In Vitro Tissue Models for Drug Discovery and Transport Studies. *Journal of Pharmaceutical Sciences*. 100(1): 59–74.
12. Allen D, Caviedes R, Cárdenas A M, Shimahara T, Segura-Aguilar J, et al. (2005). Cell Lines as In Vitro Models for Drug Screening and Toxicity Studies. *Drug Development and Industrial Pharmacy*. 31(8): 757–768.
13. Phan D T, Wang X, Craver B M, Sobrino A, Zhao D, et al. (2017). A vascularized and perfused organ-on-a-chip platform for large-scale drug screening applications. *Lab on a Chip*. 17(3): 511–520.
14. Menacho-Márquez M, & Murguía J R. (2007). Yeast on drugs: Saccharomyces cerevisiae as a tool for anticancer drug research. *Clinical and Translational Oncology*. 9(4): 221–228.
15. Khetani S R, Berger D R, Ballinger K R, Davidson M D, Lin C, et al. (2015). Micro engineered Liver Tissues for Drug Testing. *Journal of Laboratory Automation.* 20(3): 216–250.
16. Benigni R, Bassan A, & Pavan M. (2020). In silico models for genotoxicity and drug regulation. *Expert Opinion on Drug Metabolism & Toxicology*. 16(8): 651–662.
17. Piñero J, Furlong L I, & Sanz F. (2018). In silico models in drug development: where we are. *Current Opinion in Pharmacology*. 42: 111–121.
18. DI SALVO C. (2021). Limitations of In Silico Methods | proventainternational.com. *Proventa International*.
19. Luo Z, Liu Y, Zhao B, Tang M, Dong H, et al. (2013). Ex vivo and in situ approaches used to study intestinal absorption. *Journal of Pharmacological and Toxicological Methods*. 68(2): 208–216.
20. Sacan A, Ekins S, & Kortagere S. (2012). Applications and Limitations of In Silico Models in Drug Discovery. *Methods in Molecular Biology*: 87–124.
21. Umscheid C A, Margolis D J, & Grossman C E. (2011). Key Concepts of Clinical Trials: A Narrative Review. *Postgraduate Medicine*. 123(5): 194–204.
22. Ghallab A. (2013). In vitro test systems and their limitations. *EXCLI journal.* 12: 1024–1026.
23. Cutuli M A, Petronio Petronio G, Vergalito F, Magnifico I, Pietrangelo L, et al. (2019). Galleria mellonella as a consolidated in vivo model host: new developments in antibacterial strategies and novel drug testing. *Virulence*. 10(1): 527–541.

DOI:10.31579/2690-4861/322