ARCHIVES OF HELLENIC MEDICINE: ISSN 11-05-3992

REVIEW
ΑΝΑΣΚΟΠΗΣΗ

The use of animal studies in human research

Animal studies have supported our knowledge about basic mechanisms of the human body and led to the development of greatly needed forms of treatment. Yet, we cannot overlook the fact that the use of animals in research has always raised controversy on ethical and technical grounds. The use of animals in human research has long been a subject of debate in relation to its correctness and its value to research. The aim of this review is to summarize the special concerns in animal research, including the problem of animal-to-human predictability, the poor methodological standards in animal research, and the inadequate reporting of data.

ARCHIVES OF HELLENIC MEDICINE 2021, 38(6):761–765
ΑΡΧΕΙΑ ΕΛΛΗΝΙΚΗΣ ΙΑΤΡΙΚΗΣ 2021, 38(6):761–765

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Η χρήση των μελετών σε ζώα στην ανθρώπινη έρευνα

Περίληψη στο τέλος του άρθρου

Key words

Animal research
Basic research
Experimental research
Pre-clinical studies

Submitted 9.3.2021
Accepted 22.3.2021

1. INTRODUCTION

Over the past centuries, animal research has been successfully used in many areas of science, such as in basic research, and has played an important role in the development of modern medical treatments.[1,2] Even though the history of today's therapeutic armamentarium has always involved animal testing, we cannot overlook the fact that the use of basic laboratory science has always raised controversy on ethical and technical grounds, while the successful translation of promising preclinical discoveries into human studies is rare.[3] There is ongoing debate over the appropriateness and value of using animals in medical and public health research.[4] This review summarizes the special concerns in animal research, including the issue of animal-to-human predictability, the methodological flaws in animal experimentation, and the poor reporting of animal data.

2. UTILITY OF ANIMALS IN RESEARCH

Research based on animals has brought new and deeper understanding about the basic mechanisms of the human body and has provided valuable contributions to the development of great medical advances with a profound impact on such diseases as poliomyelitis and Parkinson's disease. Advances in surgical techniques and methods of treatment, including kidney and heart transplantation, were initially tested and perfected with the use of animals.[5–7] Animal studies provide a degree of environmental and genetic manipulation not often feasible in humans.[7] Experiments using animals have not only supported the development of new vaccines for the treatment of infectious diseases of public health significance, including diphtheria, tetanus, tuberculosis, poliomyelitis, and measles, but have also led to the development of greatly needed forms of treatment, such as antibacterial and antibiotic drugs.[1,8] Such studies can provide important information in terms of the pathophysiology and the causes of disease, and can disclose new targets for directed treatment. Pre-clinical studies in selected animal species are also necessary for the formulation of hypotheses that justify clinical trials. Without such studies, it would be unethical to test promising but unproven therapies in humans and it would not be necessary

to allocate valuable resources to test new treatments on humans, given that preliminary testing on animals failed to demonstrate clinical relevance.[4,9] Extensive animal testing is also required by regulatory authorities concerned with public protection, to screen new treatments for toxicity and to establish safety.[9]

3. EFFICACY AND PREDICTABILITY

While basic research is of inherent value, decades of animal experimentation for the investigation of specific diseases such as cancer and diabetes mellitus have produced little or nothing of value to humans, as encouraging results in animal studies seldom translate into successful human randomized trials with similar results.[10–12] It is estimated that around 28 billion dollars per year are invested by the United States in basic research that cannot be reproduced.[13] In addition, approximately 90% of promising discoveries examined in clinical trials fail to obtain regulatory approval, or ultimately to improve human health, because of inadequate efficacy and or unacceptable toxicity, and the limited predictive ability of preclinical studies.[12,14–16] For instance, the traditional mouse models for cancer have now been widely discredited, as human cancer cell lines are more accurate in identifying effective cancer drugs, and in practice, the traditional mouse allograft model is not predictive at all.[17–19] Similarly, the entire field of mouse immunology research is tainted by the recent discovery that, unlike humans, mice have a second, functional cervical thymus gland, which raises important questions about results from previous trials on thoracic thymectomized mice.[20] In addition, despite the use of numerous successful animal models for the treatment of traumatic brain injury, diabetes mellitus and stroke, each one has failed to translate into benefits for humans.[4,21,22] Previous studies investigating the issue of translation of published, highly promising discoveries of *in vivo* basic science into clinical applications found that only a minority of these (approximately 5–8%) were ultimately translated into an approved therapeutic method.[12,15] Consequently, all these failures of translation, expose clinical research participants to the possible danger of discoveries that fail, for reasons of safety or efficacy, and deprive them of funding for developing potential beneficial interventions.[23–27]

The concept of animal-to-human predictability is based on the hypothesis that animal studies are translatable to the human situation. Due to the complexity, this statement (or parts of it) is not always true. This can be attributed to important differences between species, ranging from genetics to physiology.[28,29] Several analyses have set out to understand why the extrapolation of results from animals to human sometimes fail. One obvious reason is the difference, not so much in organ composition and functions, but in the greater complexity of humans compared to all other animal species. It is generally accepted that laboratory animal models share some features with humans, arguably acting as excellent representations of certain human characteristics and attributes. The underlying reason for the poor translation of animal model results into human trials can be attributed to the vast anatomical, physiological, and genetic differences between them.[4,30,31] In addition, the human organism often differs dramatically from the animal species involved in pre-clinical studies with respect to the absorption, distribution and excretion of substances, and often forms very different metabolites of the same drug.[4,8] Another possible explanation is that laboratory animals are usually young, with no comorbidities, and have not been exposed to the several competing interventions that humans often receive. Finally, differences associated with the formulation, route of administration and timing of an intervention used in animal studies compared with human studies may create problems, as these factors can influence the pharmacokinetic properties of drug (e.g., absorption, distribution, etc.).[9,32]

4. BIAS AND METHODOLOGICAL LIMITATIONS

The poor quality of preclinical animal studies is widely acknowledged, and there is growing belief among scientists that an important part of the discrepancy between animal and human studies is due to the poor quality and methodological biases in animal experimentation, and the lack of adequate reporting of animal data.[4,8,9,33] Bias related to randomization, double blinding, surrogate endpoints, calculation of sample size, statistical analysis, and nonpublication of negative results continue to limit the extrapolation of animal findings to human.[4,12,34,35] For instance, an analysis of 76 animal studies published in leading journals between 1980 and 2000 showed that only around one third of highly cited animal research was finally translated to the level of human randomized trials, and only 49% was conducted according to good methodological quality.[12] In another analysis, 290 animal studies that did not use randomization or blinding were much more likely to report a treatment effect than studies that were randomized or blinded.[34] Similarly, in an analogous analysis of 4,445 animal studies in 160 meta-analyses of neurological diseases, the authors concluded that there was a possibility that most of the data were either suppressed or recast in such a way that truly negative studies would be published as positive results,

suggesting strong bias, and with selective analysis and outcome reporting bias being used as a plausible explanation.[36] In addition, a series of systematic reviews of animal studies revealed indications of selective analysis and outcome reporting bias, and also publication bias, leading to overstatement of the validity of entire bodies of medical and public health research.[10,37–41] Considering that the evidence in clinical research and public health is hierarchical, from animal studies to observational studies, randomized control trials and their secondary synthesis (e.g. systematic reviews and meta-analyses), it is possible to assume that systematic reviews often sanctify results from poor or misleading primary studies.[42,43] Umbrella reviews, which are reviews of multiple systematic reviews and meta-analyses, have been developed to assess the credibility of the evidence in an entire field, and they represent one of the highest levels of evidence synthesis today.[44–46] For instance, the risk of reporting, selection, and other inherent biases has been detected in umbrella reviews covering a very wide range of topics including nutrition,[47,48] psychiatry,[49,50] obstetrics and gynecology,[51–53] and internal medicine,[54,55] highlighting the need for cautious interpretation of primary evidence.

5. CONCLUSIONS AND RECOMMENDATIONS

In summary, knowledge gained from individual animal studies is usually incremental, each study providing knowledge that others continue to build upon for a deep understanding of physiology, at both the molecular and the macro level. To simply look at overall translation rates from single studies is an oversimplification of the scientific process and the ways in which interventional therapies have been developed. Animal studies can certainly be more beneficial in hypothesis generation than the direct prediction of the human response, considering that there is room for substantial improvement in animal research to enhance its credibility and reproducibility. It should be noted that we are in no way underplaying the importance and value of animal testing, enabling, among other things, the proliferation and testing of everyday surgical techniques, but at this point in time, we believe that is necessary to address the methodological flaws in experimental studies, such as the lack of randomization and blinding, sample sizes that do not permit valid statistical analysis, and insufficient transparency in the reporting of results.

For instance, in response to the serious deficiencies found in the conduct and reporting of animal studies the "Animal Research: Reporting In Vivo Experiments (ARRIVE 2.0) guidelines" were formulated in 2020.[56] In addition, recent attempts to improve translation from animal research also include the "co-clinical trial" in which preclinical trials explicitly parallel ongoing human phase I and II trials.[57] Likewise, a prospective registration system of animal experiments, similar to that used for clinical trials, is needed to avoid publication bias.[10] Further studies are needed to identify the prevalence of bench-to-bedside translation, to examine current trends in translation, and to identify key modifiable factors associated with successful translation of preclinical research into clinical trials, using established knowledge synthesis methods. Lastly, there is a need for preclinical researchers to push for broader dissemination of protocols, and techniques to improve the transparent reporting of preclinical studies using clear definitions and calculations. Ultimately, all these suggestions will improve the quality and the reliability of animal studies and consequently their predictive value, but they will also help researchers to distinguish truly promising therapies from the many false-positive or overstated leads.

ΠΕΡΙΛΗΨΗ

Η χρήση των μελετών σε ζώα στην ανθρώπινη έρευνα

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Αρχεία Ελληνικής Ιατρικής 2021, 38(6):761–765

Μελέτες σε ζώα έχουν υποστηρίξει τις γνώσεις μας σχετικά με τους βασικούς μηχανισμούς του ανθρώπινου οργανισμού και έχουν οδηγήσει στην ανάπτυξη πολύ αναγκαίων θεραπειών. Ωστόσο, δεν μπορεί να παραβλεφθεί το γεγονός ότι η χρήση ζώων στην έρευνα ήταν πάντα πεδίο αντιπαράθεσης για ηθικούς και τεχνικούς λόγους. Η χρησιμοποίηση ζώων στην έρευνα για τον άνθρωπο αποτελεί εδώ και πολύ καιρό αντικείμενο συζήτησης σχετικά με την

ορθότητα και την αξία του. Σκοπός αυτής της ανασκόπησης είναι η σύνοψη των ιδιαίτερων ανησυχιών στην έρευνα σε ζώα, περιλαμβανομένου του προβλήματος της προβλεψιμότητας από ζώα σε άνθρωπο, τα αδύνατα μεθοδολογικά πρότυπα της έρευνας σε ζώα, καθώς και η ανεπαρκής αναφορά των δεδομένων.

Λέξεις ευρετηρίου: Βασική έρευνα, Έρευνα σε ζώα, Πειραματική έρευνα, Προκλινικές μελέτες

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