

Use of animals in research: a brief review of legislation in Brazil

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Abstract

The use of animals for scientific purposes is a historical procedure in human civilization, but is controversial for societies concerned with the protection of animals. In Brazil, until 2008, there was no rule or law that specifically regulated animal testing. This paper discusses the use of animals in scientific experiments, considering the Brazilian Arouca Law, through the analysis of scientific articles that consider the history of experimentation in the world and in Brazil, including the regulation of the use of animals of the phylum Chordata, subphylum Vertebrata, in Brazilian research. The Arouca Law may represent an advance in Brazilian law regarding the use of animals for scientific purposes, particularly given the creation of the Ethics Committees for Animal Use in research institutions and the National Council for Animal Experimentation Control, which examine the compliance of scientific projects involving the use of such animals to applicable law.

Keywords: Bioethics. Animal technicians-Medical care. Laboratory animal science. Models, animal. Animal use alternatives. Animal experimentation.

Resumo

Utilização de animais em pesquisas: breve revisão da legislação no Brasil

A utilização de animais para fins científicos configura prática histórica na civilização humana, mas gera polêmica em sociedades preocupadas com proteção dos animais. No Brasil, até 2008, não havia norma ou lei que regulamentasse especificamente a experimentação animal. Este trabalho discute a utilização de animais em experimentos científicos, considerando o delineamento da Lei Arouca, por meio da leitura de artigos científicos que contemplam o histórico da experimentação no contexto mundial e brasileiro, incluindo a regulamentação do uso de animais do filo Chordata, subfilo Vertebrata, em pesquisas no Brasil. A Lei Arouca pode representar avanço na legislação brasileira quanto à utilização de animais para fins científicos, sobretudo pela criação das comissões de ética para uso de animais em instituições de pesquisa e do Conselho Nacional de Controle de Experimentação Animal, que examinam o cumprimento da legislação aplicável em projetos científicos que envolvem a utilização de animais.

Palavras-chave: Bioética. Técnicos em manejo de animais-Cuidados médicos. Ciência dos animais de laboratório. Modelos animais. Alternativas ao uso de animais. Experimentação animal.

Resumen

Utilización de animales en la investigación: breve revisión de la legislación en Brasil

El uso de animales para fines científicos configura una práctica histórica en la civilización humana, pero genera controversia en las sociedades preocupadas por la protección de éstos. En Brasil, hasta 2008, no había una norma o una ley que regulara la experimentación animal. Este trabajo discute acerca del uso de animales en experimentos científicos, teniendo en cuenta los lineamientos de la Ley Arouca, a partir de la lectura de artículos científicos que abordan la historia de la experimentación animal en el mundo y en el contexto brasileiro, incluyendo la regulación del uso de animales del filo Cordados, subfilo Vertebrados, en investigaciones en Brasil. La Ley Arouca puede representar un avance en la legislación brasileira con respecto al uso de estos animales para fines científicos, sobre todo por la creación de las comisiones de ética para el uso de animales (Ceua) en instituciones de investigación y del Consejo Nacional de Control de la Experimentación Animal (Concea), que son los responsables de examinar el cumplimiento de la legislación aplicable a proyectos científicos que involucran la utilización de animales.

Palabras clave: Bioética. Técnicos de animales-Atención médica. Ciencia de los animales de laboratorio. Modelos animales. Alternativas al uso de animales. Experimentación animal.

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Declararam não haver conflito de interesse.

Animal experimentation can be understood as the practice of performing interventions on live or freshly slaughtered animals in order to benefit scientific knowledge. Although developed since ancient times, the procedure is capable of offending human sensibilities and, more currently, arouses discussion among the academic community and animal protection groups^{1,2}. This confrontation even goes beyond the ethical argument and questions the real effectiveness of this teaching and research method, given the present technological and scientific advances³⁻⁵.

Thus, individuals involved with the protection of animals believe the practice is unnecessary, claiming the feasibility of using substitutive research methods, as well as the possibility of methodological errors when one wants to transfer interpretations obtained from tests on certain animal species to a different one, as in the case of humans^{4,5}. In fact, although, the use of animals in medical research has brought about success in many therapeutic interventions^{3,6,7}, detrimental effects can be observed. The drug thalidomide, for example, prescribed as a hypnotic and sedative for humans, including pregnant women, resulted in many cases of congenital malformations in children⁸. This was partly due to misinterpretation of the actual effect of thalidomide, as rodents' metabolism of the drug is different from humans' metabolism, and, as a result, numerous reports of "thalidomide babies"⁹ were observed.

In this context, it should be noted that in humans, for example, some protein isoforms are responsible for the metabolism of most commercially available anticonvulsant drugs, with the subfamilies 3A4, 2D6, 2C9, 2C19, 2E1 and 1A2¹⁰ standing out. According to Andrade et al¹¹, those same enzymes are not observed during the biotransformation of drugs in other animal species, particularly those used in biomedical research. In rats, the enzymes participating to a greater extent in this process belong to the subfamilies 1A1, 1A2, 2A1, 2B1, 2B2, 2C11, 2D1, 2E1 and 3A1, and in dogs, the CYP1A, 2B11, 2C21, 2D and 3A12 enzymes represent the greater contribution¹¹. Therefore, one can see that, depending on the species analysed, specific groups of enzymes cooperate during the biotransformation process of specific drugs, making it opportune to conjecture that each organism has its own machinery to metabolize certain drugs.

Even before this controversy, many centres of scientific research in universities resorted to animal testing in order to find cures for serious and life-threatening diseases, or to understand the

mechanism of the onset of various diseases that affect not only humans but also other living beings. Especially with regard to testing of new drugs for certain diseases, in a way the side effects observed in clinical trials can be mitigated and prevented based on observations from previous *in vivo* studies. This context highlighted the need to regulate the use of animals in scientific research in Brazil, imposing limits on this practice to eliminate acts of cruelty and maltreatment of animals used in experiments and to promote the improvement of methodological and ethical aspects of scientific studies^{12,13}.

Thus, Law 11.794, also known as Lei Arouca (Arouca Law), which regulates the procedures for the scientific use of animals¹⁴ was approved in 2008 in Brazil. With the publication of the law, commissions for the ethical use of animals (comissões de ética para uso de animais - CEUA) were created in each research institution, as well as the Conselho Nacional de Controle de Experimentação Animal - CONCEA (National Council for Animal Experimentation Control), which is now responsible for all discussions regarding the breeding and use of animals in scientific laboratories. Although the Arouca Law has spent thirteen long years to be processed, it can be said that no other Brazilian law dealt with the subject of animal experimentation so exclusively.

Considering the emergence of various theories and rules related to the use of animals in scientific research in Brazil over the years that led to the approval of the Arouca Law, this review aims to present a synthesis of global and Brazilian history of animal experimentation, as well as to assess the current regulations on the use of animals in scientific research in Brazil, based on the reading of published articles, especially in the SciELO database, and to highlight relevant and positive points of the law regarding its arguments concerning the protection of animals.

Brief history of vivisectionists practices

Dissection of animals for educational or scientific purposes has been practiced since antiquity. There are records of its origin in ancient Greece, in the experiments of Hippocrates, the "father of medicine", and Alcmeón, who in 500 BC compared animal and human organs¹⁵. In approximately the same period, physiologists, such as Herophilus (300-250 BC) and Erasistratus (350-240 BC), also resorted to animal testing in order to obtain information about the functioning of organic systems¹⁶. Aristotle

(384-322 BC) also defended the superiority of humans over animals, establishing a natural hierarchy in which beings with less reasoning ability should benefit those deemed more rational¹⁷⁻¹⁹.

Later, in Rome, Galen (130-200 AD) performed the first vivisections with an experimental purpose, inducing organic changes in animals and then evaluating the variables^{15,20}. It was through this procedure that Galen reported important structural features of the blood vessels and discovered that arteries carried blood instead of air, as it was believed for hundreds of years. Later, in 1638, William Harvey proposed the first systematic observation of dissected animals with a scientific purpose, and published the results in experimental studies on the physiology of circulation in more than eighty species^{21,22}.

Animal experimentation dates back to the times when religion and science were not clearly distinct, from each other and the advent of Judeo-Christian tradition, especially in the Middle Ages, further boosted this practice, especially when portraying animals as soulless, while prohibiting the dissection of human cadavers^{23,24}. After this period, during the Renaissance, the rise of anthropocentrism put human beings at the centre of concerns, consolidating the idea that all existing things should serve human kind, sustaining further animal testing as a standard method of scientific research and education in medicine²³.

This view prevailed in the following centuries, especially in the period of modern rationalism in the seventeenth century, when animal experimentation reached its peak. It was then that the philosopher René Descartes formulated the theory of the animal model, which considered animals as beings devoid of souls and, therefore, of the ability to feel pain, in contrast to the human species. This is the so-called “mechanistic theory” in which animals would not be more than simple machines^{23,25}.

However, in contrast to the theory created by Descartes in 1789, the philosopher and jurist Jeremy Bentham laid the foundation for moral principles and legislation currently used in ethical regulations of animal testing procedures^{17,26}. In his philosophical treatises, he encouraged society to discuss the truth of the animals lack of capacity to suffer, arguing that the ability to suffer, and not the ability to reason, must be taken into account in the way other beings are treated²⁶. This line of reasoning, was even propagated in the nineteenth century with the increased use of animals in scientific research laboratories, a time when the first animal protection organisations came into being, extending to the present day.

In 1822, the British Anticruelty Act was instituted to nullify acts of animal torture, but it was only applicable to large domestic animals²⁷. In the year 1824, the Society for the Prevention of Cruelty to Animals was established in England, which was an important step for the foundation of similar societies in other countries, including Germany, Belgium, Austria, the Netherlands and United States²⁷. However, the first dedicated law to regulate the use of animals in research^{17,27} only appeared in England in 1876.

It is important to remember that about this period (1858-1859), Charles Darwin gave great impetus to scientific research around the world with the publication of “The Origin of Species”, which depicts the interaction between different species during the evolutionary process^{17,28}. This event certainly reinforced the possibility of considering for the human species the information obtained in tests with other animal species.

In the early twentieth century, specifically in 1909, the first publication regarding the ethical use of animals in experiments was proposed by the American Medical Association^{17,27}. Fifty years later, a major step aimed at the establishment of ethics in animal research was made when the zoologist William Russell and the microbiologist Rex Burch established the three “R’s” of animal research: *replace, reduce and refine*^{17,27}. Later, Peter Singer²⁸ suggested that the suffering of each species be analysed and compared to the one experienced by a member of another species, although he admitted that this comparison is not quite accurate.

During this period, there was growing concern regarding the ethical and social impact of scientific research involving animals. Thus, the United Nations Educational, Scientific and Cultural Organization (UNESCO), while holding an event in Brussels in 1978, signed the *Universal Declaration of Animal Rights*, leading society to discuss, even more intensely, the need for animal experiments²⁹.

On 8th May 1979, Brazil passed the Law 6638 to regulate animal testing in higher education institutions, determining that these studies could be carried out provided that the animals were not kept in conditions that cause them suffering³⁰. In 2008, the *Projeto de Lei 1153/1995* (Bill 1153/1995) was approved, authored by the former Deputy Sérgio Arouca³¹, transforming into the *Lei Ordinária 11794/2008* (Common law 11794/2008)¹⁴, which repealed the previous law and currently regulates the breeding and use of animals in educational activities and scientific research throughout Brazil.

Animal protection in Brazil

In Brazil, the first document that dealt with animal protection, was dated 6th October 1886, a time when slavery was to be abolished in the country^{32,33}, was part of the Código de Posturas do Município de São Paulo (São Paulo Code of Behaviour). Article 220 of the code deterred acts of ill-treatment, such as barbarous and inordinate forms of punishing animals, as used by coachmen, blacksmiths, grooms or horse-drawn vehicle drivers^{32,33}, since, until then, in the early Republican era and with the dissemination of vehicles powered by animal traction, in the absence of legislation, these unpunished acts of abuse and maltreatment were common³³.

In 1916, Article 47 of the Código Civil (Civil Code)³⁴ indicated that animals were considered property objects. However, it was succeeded by laws dealing with the protection of animals in a more concrete way, such as Decree 16590/1924³⁵, which forbade any entertainment developed at the expense of animals acts of cruelty and ill-treatment, such as cattle races or bird fights in public places of entertainment³⁶. Subsequently, during the government of Getulio Vargas, Decree 24,645/1934³⁷ was promulgated, determining the protection by the state of all animals in the country and, in its Article 3, defining as conducts of abuse acts of cruelty, violence and excessive work, maintenance of the animal in unhygienic conditions and abandonment^{36,37}. Considering the absence of a specific law to regulate the practice of vivisection in Brazil, this decree was also used as a reference standard for animal experiments³⁸.

Already in 1941, great progress was made with Decree-Law-3688³⁹, also known as the Lei das Contravenções Penais (Law of Criminal Offenses), which in article 64 established the penalty of simple imprisonment for acts of animal cruelty, regardless of educational or scientific purpose of the act¹³. In that year, it was found necessary to also criminalize the conduct involving animals in scientific research laboratories or academic environments, and therefore the practice of painful or cruel experiments on animals, in addition to those acts performed in public places, was subsequently elevated to a criminal offense.

Subsequently, other laws have been passed, such as the Lei de Proteção à Fauna (Lei 5197/1967) (Wildlife Protection Law - Law 5197/1967)⁴⁰ and the Código de Pesca (Decreto-Lei 221/1967) (Fisheries Code Decree-Law 221/1967)⁴¹, but none specifically addressed the topic “animal experimentation

for educational or scientific purpose”. Given this situation, the *Projeto de Lei 1507* [Bill 1507]⁴² was introduced in August 1973 by Deputy Peixoto Filho, resulting later, in 1979, the enactment of *Lei 6638* [Law 6638]³⁰, which deals with permissions and procedures for the practice of animal experimentation nationally and requires that animal facilities, experimentation centres and demonstrations with animals must be registered with the competent organization.

This law was the first to establish, in Brazil, standards directly applicable to the practice of animal experimentation with didactic and scientific purpose, authorizing the exceptional practice of vivisection in higher education institutions throughout Brazil. However, despite Article 6 establishing the period of 90 days for regulation of this law by the executive branch, the law never received proper regulation. Consequently, no legal authority or competent organisation was appointed as responsible for monitoring compliance with its rules and for registering institutions and professionals dedicated to the use and breeding of animals for didactic and scientific purpose. Similarly, there was no specification as to the environmental conditions or organisations responsible for the inspection of animal facilities and research laboratories that conducted animal husbandry³⁰.

Even so, the promulgation of the law has made considerable progress with the establishment of norms used for the protection of animals used in scientific research or university centres, as Article 3 prohibits animal experiments without the use of anaesthesia or acclimatization of animals to vivarium for a period of fifteen days. Furthermore, according to Article 4, the didactic and scientific procedures would only be allowed by ensuring special animal care throughout the completion of the experimental protocol³⁰.

In 1998, another advance toward the outcome of the ethical implications of the use of animals in teaching and scientific experiments took place with the introduction of *Lei de Crimes Ambientais* (Lei 9605/1998) (Environmental Crimes Law - Law 9605/1998)⁴³, which reordered the Brazilian environmental legislation in relation to offenses and punishments. This law increased the severity of the penalty for the practice of ill-treating animals in general or for performing painful or cruel experiments on live animals, even for educational or scientific purposes, when substitute resources exist.

However, until October 2008 there was no law that dealt in more detail with the subject of experimenting on animals, demanding educational

and research institutions to register with the state superintendence of the Instituto Brasileiro do Meio Ambiente e dos Recursos Naturais Renováveis – IBAMA³⁸ (Brazilian Institute of Environment and Renewable Natural Resources).

Lei Arouca (Arouca law) and the protection of animals - current landscape and observations

As stated, the Lei Arouca (Arouca Law) came into being in a scenario in which there was intensified use of animals for scientific research, while there were no regulations aimed specifically at vivisectionist practices on animals for educational or scientific purpose. Although starting late compared to other developed countries, this law is the result of the increased debates, since the 1990s, regarding the use of animals in scientific research in Brazil.

Despite offering more specific legislation on the subject and also creating organisations responsible for monitoring compliance with its rules, the promulgation of the Arouca Law caused even more controversial clashes between the scientific community and animal protection groups, as it did not correspond to the expected abolition of the use of animals in scientific practices⁴⁴.

However, it is necessary to consider that Brazil is growing as a country that conducts scientific research. Therefore, it is remarkable that, until 2008, this nation did not yet have a law that specifically regulated animal research. The Arouca Law, therefore, brought the country to another level, to that of nations seeking to protect animals used in research.

In ancient times animals, in general, were regarded as being unable to suffer in the face of environmental adversity, while vivisections were practiced without legislation that regulate them and thereby limit the number of animals and their suffering. The present day is defined by questions regarding the progress made as a result of the publication of the Arouca Law. Along this line, some authors⁴⁵ believe that the law is a step backwards, saying that instead of contributing to the defence of animals and awareness of humanity, it creates opportunities for the realization of numerous vivisectionists practices, contributing to the increased recurrence of scientific research on animals.

In fact, compared to the old Law 6638, Arouca Law, in Article 1, extends the permission of vivisectionist practices on animals in institutions focused on technical secondary biomedical education, in addition to those of higher education¹⁴. However,

there is a need to deepen the knowledge of the standardization of the law to understand what has evolved in relation to respect for animals, considering that it aims to care and limit the number of animals used in experimental protocols.

Firstly, one of the ways of ensuring compliance with the standards for vivisectionist practices on animals is Article 13, which states that all institutions responsible for creating or using animals for teaching and research should be legally established in the country, have accreditation with the Concea and must create one or more Ceua¹⁴.

Similarly, Article 5 attributes to the CONCEA the task of formulating rules related to the use of animals for the purposes of teaching and scientific research and ensuring institutions' compliance. The Concea should also keep up to date the record of teaching and research procedures performed or in progress in the country, as well as the records of the researchers, from information sent by Ceuas. The same Article 5 states that the Conselho Nacional (National Council) should further determine and revise technical standards for the installation, operation and working conditions of breeding centres, animal facilities and animal experimentation laboratories, recommending the appropriate conditions for animal maintenance in such teaching and research environments. In addition, the functions of Concea include monitoring and evaluating the introduction of alternative techniques that replace the use of animals in teaching and research, possibly in an attempt to control and veto the conducting of experimental and teaching protocols that can be replaced by study methods that do not utilise animals¹⁴.

The duties of Ceua are well explained in Article 10, which determines the prior examination by the committees of all procedures to be performed in experimental protocols for scientific projects developed in the associated institution, and seeks verification of research project compliance with applicable legislation. With this objective, Ceua meetings are held periodically, that even include the participation of representatives from animal protection organisations legally established in Brazil¹⁴.

The Arouca Law also stipulates, under Article 14 (paragraph 3), that teaching practices should, whenever possible, be photographed, filmed or recorded, to allow their reproduction for illustrating future educational and research practices, avoiding the unnecessary repetition of didactic procedures with animals¹⁴. Cell cultures, computer simulations and mathematical modelling, among others, are

substitutive methods for the use of animals for educational or scientific purposes.

In this way, it verifies adherence to the “replace” and “reduce” principles of the three experimental “R’s”^{17,27}, which aim at establishing the use of substitute methods and reducing the number of animals in research and teaching. It should be noted, however, that the use of replacement methods is not always possible, even with the technological sophistication of today, because there are still no ways to mimic the complexity of interactions between cells, tissues and organs that occur in living organisms, human beings and animals, which instigates conducting *in vivo* studies in order to facilitate understanding of the functioning of the human organism, and consequently, development of new treatments for numerous lethal and limiting medical conditions.

Finally, in relation to the care provided to animals in scientific research, Article 14 of the Arouca Law aims to ensure attention for their well-being throughout the experimental protocol of scientific interventions¹⁴, following the principle “refinement” of Russell and Burch^{17,27}. In fact, it advocates: the banned re-use of animals after obtaining the goal of the research (Article 14, paragraph 8); recurrent sedation techniques, analgesia or anaesthesia appropriate to the animal species when the experiments cause pain or distress, which, in turn, require specific authorization from Ceua (Article 14, paragraphs 5 and 6); and possible restriction of highly aggressive procedures (Article 15)¹⁴. In addition, the sacrifice of animals, when necessary, should also be appropriate to the species and follow ethical and acceptable methodological standards (Article 14,

paragraph 1). Also in this issue, there are specific guidelines⁴⁶ that assist the researcher in selecting the best form of euthanasia for animals used in scientific experiments.

Final considerations

Animal experimentation, especially those focused on scientific research, should not necessarily be banned, since the progress made in the knowledge of physiology, pharmacology and pathology would not have been possible without *in vivo* studies. In this context, the enactment of the Arouca Law becomes beneficial to the Brazilian scientific research, coupled with the protection of animals, as it enables, with the creation of organisations such as CEUAS and CONCEA, the exceptional use of animals in scientific studies when it has a positive impact for the world population and when it is done consciously and methodically free from mistreatment. Therefore, the Arouca Law is not detrimental to the protection of animals.

Brazilian law has advanced, albeit slowly, regarding the matter of regulating the use of animals in educational or scientific practices, and certainly the Arouca Law can be considered part of this advance. In fact, the terms of the current legislation for the breeding and use of animals aimed at teaching and research imposes limits on the practice, taking into account, as much as possible, the protection of animals, as it recommends the planning of experiments in order to use the smallest possible number of animals and to avoid unnecessary stress, pain or suffering.

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Mariana Vasconcelos Guimarães developed the concept and the initial draft, which were edited by José da Cruz Ednésio. Lea Maria Bezerra de Menezes made the final version.

