

# Knowledge of elementary and high school students about experimental animal research

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#### **Abstract**

Experimental research with the use of animal models to represent a specific reality is a target of criticism by the population. This study analyzed the knowledge of elementary and high school students on experimental/animal research methods, official ethical guidelines/laws, and regulatory institutions. In total, 35 students answered an informative questionnaire with objective questions about the subject. Only 18 students (Group 1), attended a lecture on the subject. The questionnaire was reapplied to all 35 students. The analysis of the first questionnaire round showed rejection by 51.4% of the students toward the methods used in experimental research. Significant changes in answer patterns between the first and second evaluations were observed, with a decrease in the number of students who strongly disagree with the methods used in experimental research in Group 1 (38.8%) compared to Group 2 (88.2%). These data suggest that educational activities could increase the acceptance of experimental research by the community.

Keywords: Animal experimentation. Drug development. Ethics committees. Vaccines.

#### Resumo

Conhecimento de alunos dos ensinos fundamental e médio sobre pesquisa experimental em animais A pesquisa experimental com uso de modelos animais para representar uma realidade específica é alvo de críticas por parte da população. Este estudo analisou o conhecimento de alunos do ensino fundamental e médio sobre métodos de pesquisa experimental/animal, diretrizes/leis éticas oficiais e instituições reguladoras. Ao todo, 35 alunos responderam a um questionário informativo com perguntas objetivas sobre o assunto. Apenas 18 alunos (Grupo 1) assistiram a uma palestra sobre o tema. O questionário foi aplicado novamente aos 35 alunos. A análise do primeiro questionário mostrou rejeição por 51,4% dos estudantes em relação aos métodos utilizados na pesquisa experimental. Observaram-se mudanças significativas nos padrões de resposta entre a primeira e a segunda avaliação, com diminuição do número de alunos que discordam totalmente dos métodos utilizados na pesquisa experimental no Grupo 1 (38,8%) em relação ao Grupo 2 (88,2%). Esses dados sugerem que atividades educativas podem aumentar a aceitação da pesquisa experimental pela comunidade.

Palavras-chave: Experimentação animal. Desenvolvimento de medicamentos. Comissão de ética. Vacinas.

### Resumen

Conocimiento de los alumnos de la primaria y la secundaria sobre investigación experimental en animales La investigación experimental en animales para representar una realidad específica es criticada por la población. Este estudio analizó el conocimiento de los alumnos de la primaria y la secundaria sobre los métodos de investigación experimental/animal, los lineamientos/leyes éticas oficiales e instituciones que reglamentan. Un cuestionario con preguntas objetivas sobre el tema fue respondido por 35 alumnos. Solo 18 alumnos (Grupo 1) asistieron a una conferencia sobre el tema. El cuestionario se aplicó nuevamente a 35 alumnos. El primer cuestionario mostró que el 51,4% de los alumnos rechazaban los métodos utilizados en la investigación experimental. Hubo cambios significativos en las respuestas entre la primera y la segunda evaluación, con menor número de alumnos que estaban totalmente en desacuerdo con los métodos de la investigación experimental en el Grupo 1 (38,8%) en comparación con el Grupo 2 (88,2%). Por tanto, las actividades educativas pueden incrementar la aceptación comunitaria de la investigación experimental.

Palabras clave: Experimentación animal. Desarrollo de medicamentos. Comités de ética. Vacunas.

The authors declare no conflict of interest. Approval CEP/CAAE 25047419.9.0000.5505 Experimental research is the use of several models to represent a specific reality, so biological, physical, chemical, and social phenomena can be studied and research findings can be applied to other species and multiple contexts<sup>1</sup>. Experimental research is associated with concerns about protecting human health against the possible toxicity of new compounds, with restrictions on experiments with humans stipulated in the *Nuremberg Code* and the *Declaration of Helsinki*.

The selection criterion for experimental models is based mainly on the metabolic and physiological similarities of each animal with human beings<sup>2</sup>. This sort of research is adopted in basic or applied scientific research laboratories, and for pharmaceutical and cosmetics industries. Modern scientific knowledge is a direct result of this kind of experimentation<sup>1</sup>. Experimental research takes into account ethical principles, the relevance of the study, and the potential results generated as well as its social impact.

Although this type of experiment is conducted since the 5th century BC with Hippocrates, debates about ethical issues only started in the early 1800s<sup>3</sup>. England was the first country to create a law against animal cruelty<sup>1</sup>. Animal protection laws spread after this, and animal protection societies were created around the world<sup>3</sup>.

Discussions about ethics in animal testing started in the 1930s, followed by the Brazilian law on animal experiments from 2008. The law regulates activities related to the use of animals for teaching and research, proposes alternative techniques, and stipulates the improvement of laboratories and animal facilities <sup>4</sup>. The Brazilian legislation on animal testing also includes the existence of regulatory institutions, such as the National Council for the Control of Animal Experimentation (CONCEA), Register of Institutions for the Scientific Use of Animals (CIUCA), and Commission for Ethics in the Use of Animals (CEUA).

Experimental research in animals is a constant target of criticism by the population, which associates the practice with animal suffering, sometimes disregarding measures taken to ensure animal well-being. Scientists must follow ethical guidelines and apply concepts such as the 3Rs and the 5 freedoms. The 3Rs refer to

replacement (replacement of animal models for other models), reduction (reducing the number of animals used), and refinement (sophistication of study methods) <sup>5</sup>.

The concept of 5 freedoms considers the needs and rights of animals, ensuring nutrition, environment, and physical and mental health, and demanding freedom from hunger and thirst, discomfort, pain, injuries and disease, to express normal behavior, and fear and distress. Measures such as keeping temperature, humidity and noises control, and cage size, which must be proportional to the animal's size are some of the measures to ensure animal comfort. Toys and accessories are used to ease the stress of caged animals, a procedure named environmental enrichment.

This resistance to animal experimentation may also result from the circulation of information from unreliable sources and the lack of disclosure about how experimental research is conducted and knowledge regarding the legislation that guarantees ethics in the scientific progress achieved with animal experimentation.

Given this context, this study aimed to evaluate the knowledge and opinion of elementary and high school-aged students about the theme "animal experimentation and its regulations in Brazil" by assessing the impact of educational interventions based on the topic on the development of a more favorable positioning concerning experimental research for the development of new drugs and vaccines.

# Method

This was a controlled and prospective experimental study, with a random sample and masking of the participants. The participants (if at least 18 years old) or legal responsible (if under 18 years old) agreed to the research by signing an informed consent form; individuals under 18 years of age signed another specific term. Students were contacted through the schools, which received all necessary information about the project, including the consent form and the online questionnaire link.

The group selected to participate in the project included 35 elementary and high school-aged students from a private school in São Paulo/SP,

from March to August 2022. The initial sample was randomized into 2 groups, one that was invited to attend a lecture (Group 1 (G1): n=18) and another that did not (Group 2 (G2): n=17).

The research instrument was a written and informative questionnaire with objective questions about experimental research with animal models, the existence of specific standards and guidelines, in addition to regulatory organizations. To ensure anonymity, only the age, gender, and current year at school were collected.

At first, all students were asked to answer the questionnaire, aiming to evaluate their knowledge/opinion but also to inform them. G1 was invited to attend an online lecture on the use of animals in experimentation, focusing on aspects of medication/vaccine production, animal rights, and protective societies. The lecture discussed the scientific advances obtained through this method, the care taken with the animals used, and the Brazilian legislation on this type of research, including the penalties applied to researchers who do not comply with the law.

Approximately 1 month after the primary intervention, the questionnaire was reapplied to groups 1 and 2. In this second questionnaire, students informed whether they had or not attended the lecture and if they would like to visit an experimental research laboratory and animal testingfacilities.

# Statistical analysis

The categorical data of gender and answers to the questionnaire were described in absolute (n) and relative (%) frequency, and continuous data (age) in the form of mean±standard deviation. For the comparison between the independent samples, the unpaired Student's t-test was used. The Fisher test was used to compare categorical data. The significance level adopted was p < 0.05. The Graphpad Prisma software was used for the analyses.

# **Results**

# Global sample

The mean age of the sample was 16 years (ranging from 14 to 18 years old), with 13 male and

22 female students, attending elementary school (n=20) or high school (n=15) in 2022.

The analyses of the answers from the entire group in the first questionnaire show that most students believed to know the concept of experimental research (Table 1). Regarding their opinion about the 3 phases of the experimental research (1. studies in computational models and/or cultivated cells, 2. animal models, 3. human beings), almost half agreed with the 3 phases; a minority disagreed with the 3 phases; and the others agreed partially. Those that partially agreed were mostly in favor of phase 1, followed by phases 3 (human beings) and 2 (animal models).

Regarding the disagreement with the scientific method phases, 42.8% of participants believe that using animal models deprives the animals of freedom; 37.1% believe that animal models harm the animals; 25.7% believe that animal models cannot predict what will happen to human beings; 14.2% would use medication, vaccines, and cosmetics that were not tested for safety and effectivity in computers, cells, and animals; 5.7% believe that isolated cells cannot predict what will happen with an animal model or human beings; and 2.8% believe that experimental research is too expensive do not trust the computational models or had other justifications.

Most students knew that it is illegal to do any experimental research without a license from an independent committee specialized in ethics in research, with composed of members from several professions and civil society. Most also knew that experimental research in humans is regulated and controlled by ethics committees worldwide to forbid the exposure of humans to risks without potential benefits previously obtained with laboratory and animal testing. Most were aware of the laws and regulatory organizations for the use of animals in research and that punishments, penalties, and bans are applicable to researchers who violate experimental research laws.

Alternatively, a minority knew that Brazil has one of the most advanced legislations for the protection and well-being of animals in experimental research since 2008 (Law 11,794/2008, known as "Arouca" Law). Likewise, only a third of the participants knew that the

animals used in experimental research are housed in special environments called animal facilities, which abide to all the requirements to decrease animal stress and guarantee well-being.

Although most knew that any procedures considered painful or distressful should be done

under anesthesia, only a third knew that after an invasive procedure, such as surgery, animals must be painlessly euthanized, under anesthesia. Interestingly, most were interested in the research field and would like to know more about the subject.

Table 1. Answers to the first questionnaire from all participants (n=35)

Questions	1 <sup>st</sup> questionnaire n=35
1. Do you know what experimental research is?	22 (62.8%)
2. Regarding the phases of experimental research:  * I agree with the three phases  * I partially agree  * I disagree with the three phases	17 (48.57%) 16 (45.7%) 2 (5.71%)
2.1. I partially agree with (more than 1 possible answer): Phase 1: computational models/cells Phase 2: animal models Phase 3: human beings	15 (42.85%) 1 (2.85%) 6 (17.14%)
4. Do you know that it is prohibited to execute experimental research without a license from ethics committees?	21 (60%)
5. Have you been informed of the fact that experimental research in humans is regulated and controlled by ethics committees?	21 (60%)
6. Have you heard about the existence of laws and regulatory institutions for the use of animals in research?	23 (65.71%)
7. Are you aware of the existence of the Arouca Law?	4 (11.42%)
8. Do you know that the violation of rules and laws of experimental research can lead to punishment for researchers?	29 (82.85%)
9. Are you aware of the existence of animal facilities?	11 (31.42%)
10. Do you know that anesthesia is used for painful or distressing procedures?	25 (71.42%)
11. Have you heard about the sacrifice of animals after invasive procedures, under anesthesia?	13 (37.14%)
12. Do you have any interest in the research field?	26 (74.28%)
13. Would you like to know more about the subject?	30 (85.7%)

# Group 2

# Isolated effect of the questionnaire

After the second questionnaire was applied, the analysis of Group 2 (Table 2), not submitted to the intervention with the lecture, showed a significant difference with an increase in the number of students who believe to know the concept of

experimental research; and a shift from those who totally agreed with the scientific method and those who totally disagreed when compared to the group that partially agreed with the method.

No significant change was observed regarding the phases of the experimental research among those who partially agreed, though an increase in the reasons for disagreement with the phases of the scientific method was observed, except for the reasons "I do not trust the computational models," "using animal models deprives the

animals of freedom," and "experimental research is too expensive."

**Table 2.** Answers to the first versus second questionnaire from Group 2 (n=17), for the assessment of the isolated effect of the questionnaire

Group 2: questionnaire effect – did not attend the lecture on experimental research	1 <sup>st</sup> questionnaire n=17	2 <sup>nd</sup> questionnaire n=17	p (Fisher's test)
1. Do you know what experimental research is?	9 (52.9%)	16 (94.1%)	0.0167
Regarding the phases of experimental research:     I agree with the three phases     I partially agree     I disagree with the three phases	8 (47%) 7 (41.17%) 2 (11.7%)	2 (11.7%) 15 (88.2%) 0 (0%)	0.0104 (partially agree vs. other groups)
2.1. I partially agree with: Phase 1: computational/cell models Phase 2: animal models Phase 3: human beings	7 (41.17%) 0 (0%) 2 (11.7%)	15 (88.2%) 1 (5.88%) 6 (35.3%)	-
4. Do you know that it is prohibited to conduct experimental research without a license from ethics committees?	10 (58.82%)	11 (64.7%)	-
5. Have you been informed of the fact that experimental research on humans is regulated and controlled by ethics committees?	8 (47%)	7 (41.17%)	-
6. Have you heard about the existence of laws and regulatory institutions for the use of animals in research?	11 (64.7%)	9 (52.9%)	-
7. Are you aware of the existence of the Arouca Law?	2 (11.7%)	0 (0%)	-
8. Do you know that the violation of rules and laws for experimental research can lead to punishment for researchers?	13 (76.47%)	11 (64.7%)	-
9. Are you aware of the existence of animal facilities?	7 (41.17%)	9 (52.9%)	-
10. Do you know that anesthesia is used for painful or distressing procedures?	13 (76.47%)	14 (82.35%)	-
11. Have you heard about the sacrifice of animals after invasive procedures. under anesthesia?	6 (35.3%)	7 (41.17%)	-
12. Do you have any interest in the research field?	12 (70.58%)	15 (88.23%)	-
13. Would you like to know more about the subject?	15 (88.23%)	16 (94.1%)	-
14. After answering the first questionnaire, did you do any research on this subject?	-	8 (47%)	-

# Group 1

# Combined effect of the questionnaire and the lecture

Group 1 also attended a lecture. To isolate its effect, the answers from G1 in the second questionnaire were compared to the answers of the

group also in the first questionnaire (Table 3). There was a significant difference regarding 3 questions.

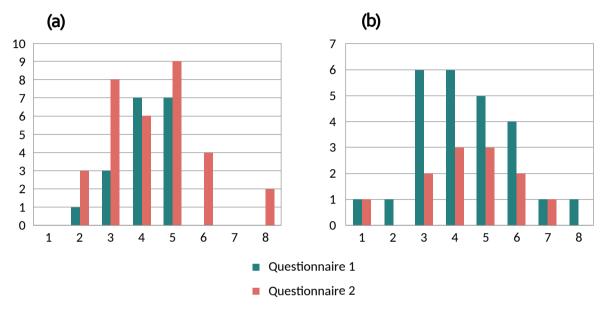
In the comparison with G2, G1 showed a shift from those who partially agreed with the scientific method to those who totally agreed with the method. Among those who partially agreed, no significant change regarding the phases of the experimental research existed.

Regarding the disagreement with the phases of the scientific method, the comparison between both questionnaire rounds showed expressive changes (Figure 1), except for "I do not trust computational models". Moreover, G1 was more aware of the laws and regulatory organizations for the use of animals in research. G1 also had more students aware that Brazil has one of the most advanced legislations for the protection and well-being of animals in experimental research since 2008.

Table 3. Answers to the first versus second questionnaire from Group 1 vs. 2 (n=18) for the assessment of the lecture effect

Question versus group	2 <sup>nd</sup> questionnaire Group 1 (lecture) n=18	2 <sup>nd</sup> questionnaire Group 2 (no lecture) n=17	p (Fisher's test)
1. Do you know what experimental research is?	17 (94.4%)	16 (94.1%)	-
<ul> <li>2. Regarding the phases of experimental research:</li> <li>* I agree with the three phases</li> <li>* I partially agree</li> <li>* I disagree with the three phases</li> </ul>	11 (61.1%) 7 (38.8%) 0 (0%)	2 (11.7%) 15 (88.2%) 0 (0%)	0.0045 (agree with the 3 phases vs. partially agree)
2.1.I partially agree with: Phase 1: computational/cell models Phase 2: animal models Phase 3: human beings	6 (33.3%) 2 (11.1%) 2 (11.1%)	15 (88.2%) 1 (5.88%) 6 (35.3%)	-
4. Do you know that it is prohibited to conduct experimental research without a license from ethics committees?	12 (66.6%)	11 (64.7%)	-
5. Have you been informed of the fact that experimental research on humans is regulated and controlled by ethics committees?	11 (61.1%)	7 (41.17%)	-
6. Have you heard about the existence of laws and regulatory institutions for the use of animals in research?	16 (88.8%)	9 (52.9%)	0.0275
7 Are you aware of the existence of the Arouca Law?	6 (33.3%)	0 (0%)	0.0191
8. Do you know that the violation of rules and laws for experimental research can lead to punishment for researchers?	16 (88.8%)	11 (64.7%)	-
9. Are you aware of the existence of animal facilities?	11 (61.1%)	9 (52.9%)	-
10. Do you know that anesthesia is used for painful or distressing procedures?	14 (77.7%)	14 (82.35%)	-
11. Have you heard about the sacrifice of animals after invasive procedures under anesthesia?	10 (55.5%)	7 (41.17%)	<u>-</u>
12. Do you have any interest in the research field?	14 (77.7%)	15 (88.23%)	-
13. Would you like to know more about the subject?	16 (88.8%)	16 (94.1%)	-
14. After answering the first questionnaire, did you do any research on this subject?	8 (44.4%)	8 (47%)	-

**Figure 1.** Answers to question 3 (If you disagree with any phase of experimental research. why? Select as many as you want). (a). Group 2 (without intervention) in the 1st vs. 2nd questionnaire (n=17); (b). Group 1 (with interventional lecture) in the 1st vs. 2nd questionnaire (n=18). Reasons: 1. I do not trust computational models; 2. Isolated cells cannot predict what will happen in animal models or human beings; 3. Animal models cannot predict what will happen to human beings; 4. The use of animal models deprives animals of freedom; 5. The use of animal models harms the animals; 6. I would use medications, vaccines, and cosmetics that were not previously tested in computers, cells, and animals regarding safety and effectiveness; 7. Experimental research is too expensive; 8. Others.



## Discussion

Our results indicated that, in general, the students from private elementary and high school-level Brazilian school believe to know the method of experimental research and agree with its phases. However, they feel greater restrictions toward experimental research with animal models than with humans. Some concepts about experimental research with animals that may ground those restrictions were detected, ranging from the view that animals suffer and are deprived of freedom, to the belief that animal models cannot predict what will happen to human beings.

Animal models enable the study of specific processes with the aim of improving human health conditions. The use of animal models complies with the *Declaration of Helsinki* in the sense that medical research in human beings must be preceded by laboratory experiments <sup>1</sup>.

Research normally evolves from cell cultures to tests in animals and then tests in humans.

still depending on animal experimentation for development or results validation. However, the risks and benefits must be balanced to reach a reasonable course of action <sup>7</sup>. Public support for research with animal models thus depends on the demonstration of the potential benefits of a research combined with experimental techniques and integral application of the 3Rs <sup>5</sup>. Regarding animal freedom, it is fundamental to disclose to society that laboratory animals would not survive in their natural habitat if they were to be released into nature <sup>8</sup>.

Most students in our sample knew about the need of a license to do research, particularly with humans, but also with animals; about anesthesia use to prevent discomfort/stress, and the laws/organizations available as well as the penalties/fines. The students showed great interest in research, particularly in the experimental area. Results regarding unawareness about how advanced the Brazilian legislation of animal protection in research is, the existence of animal facilities

specialized in animal well-being, and the procedures for euthanasia to avoid suffering were also high.

Standards for the didactic-scientific practice of animal vivisection exist in Brazil since 1979, with Law 6,638, which was later replaced in 2008 <sup>3</sup>. Supervision stimulates society to adopt laws based on animal bioethics, which will reflect in respect toward animals used in research <sup>4</sup>. The Brazilian Environmental Act (1998) reinforced this idea by determining punishment for the use of animals in experiments when alternative methods are available <sup>9</sup>.

The support of society for the use of animals in medical research was based on the belief that there were no suitable alternatives to it?. Interestingly, society supports the use of animals in research that contributes to treatments and reduces human suffering but not support in basic research?. Our results pointed to positive effects both from the informative questionnaire and from the lecture, even if the effect happened in different questions.

While the questionnaire led to an increase in students who partially agree with the three phases of an experimental study, the lecture led to an increase in those who totally agreed, associated with a reduction of questionable concepts. Nevertheless, the lecture allowed a better understanding of the Brazilian legislation to protect animals in research.

The small sample size and the impossibility to impede the students who attended the lecture

talking about it with their colleagues who did not attend are limitations of this study. Since our sample was restricted to elementary and high school-aged students, further studies with students from other educational levels would be necessary.

# Final considerations

Research using animals faces resistance from society, stronger than that toward research with humans. Students in the sample of this study associated the experimental research methods with suffering and lack of freedom for animals. However, most of them would like to know more about the subject. Which demonstrates opportunities for educational initiatives to be implemented by Brazilian researchers. In this scenario, bioethical legislation and the work of ethics committees could be more publicized.

The interventions, in the form of an informative questionnaire and lecture, lead to a more favorable position towards experimental research. These results illustrated that access to trustful information is crucial for a better understanding of the scientific process and a more favorable position regarding experimental research for the development of medications and vaccines. This position can be positively reinforced by a greater disclosure of scientific advances based on experimental models.

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#### **Participation of the authors**

Giovanna Corsino Marchioni collected data, participated in the theoretical and practical steps of the study and in the writing of the article. Monica Levy Andersen designed the study, was responsible for giving the lecture about experimental research for the participants and was involved in the writing of the article. Helga Cristina Almeida da Silva designed the study and was involved in the writing of the article.

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