



An Exploratory Study of Students' Perceptions on the Use of Animals in Medical and Veterinary Medical Undergraduate Education

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Abstract

Animals are frequently utilized as a teaching-learning tool in multiple educational settings. It is, therefore, important to understand what students think about this topic, in particular medical and veterinary students as “life caregivers” and competent people for a dynamic and responsible social intervention. In this context, this research aims to characterize and disseminate a set of issues related to animal welfare/wellbeing in higher education in the North of Portugal, particularly as regards the teaching of students of the Integrated Master in Medicine (MIM) and Veterinary Medicine (MIMV). After ethical approval, a survey was delivered on paper to 180 undergraduate MIM (n = 100) and MIMV (n = 80) students. After collecting 139 questionnaires partially or fully completed, with varying response rates for each question, it was concluded that most of the students consider that animal experimentation is ethically acceptable when the benefits balance the harms and assuming that refinement of animal procedures is warranted; they also agree to the establishment, maintenance and performance of animal procedures solely for educational purposes as a way of ensuring optimal acquisition of theoretical knowledge, attitudes and behaviors and technical skills. Nevertheless, it is the responsibility of teachers to explore and implement pedagogical methodologies that are equally effective but more humane and compassionate towards sentient living beings.

Keywords Animal welfare/wellbeing · Ethics · Higher education · Medicine · Veterinary medicine · Northern Portugal

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Introduction

In the context of life and health sciences education, animals are frequently utilized as educational tools, particularly in the undergraduate training of Medicine and Veterinary Medicine students. This usage ranges from benign observation to the dissection of cadavers, or even invasive procedures performed on living animals. However, the pedagogical paradigm asserting that *in vivo* studies lead to more significant learning has come under ethical and moral scrutiny (Andreoli et al., 2023). Considering that veterinary and medical professionals bear a greater responsibility in supervising and promoting respect for animal life and their welfare/wellbeing, are the current educational methods, that involve the use of animals, consistent with the concomitant teaching of the intrinsic value of life (Sathyanarayana, 2013; Baptista, 2019)? Is harmful animal use an acceptable methodology to be pursued for learning and skills development (Lairmore & Ilkiw, 2015)? There is also a growing awareness that subjecting animals to harmful procedures may undermine students' sensitivity, as well as their moral or religious values (Capaldo, 2004). Indeed, an increasing number of undergraduates is questioning the system by refusing to dissect animals or perform other invasive procedures, if these practices challenge their ethical values towards animals (Knight, 2014; Baptista, 2019). Additionally, many teachers have already acknowledged that students can learn equally effectively through the use of multiple non-animal alternatives (e.g. Knight, 2014), emphasizing the fact that there is a continuous need for careful and consensual reflection regarding the use of animals in educational programs that lead to the graduation of Medicine and Veterinary Medicine students. Currently, there is a generalized consensus that reducing the overall number of animals utilized as teaching-learning tool is both necessary and ethically justified. However, opinions vary on whether their use can or should be completely eliminated (Carroll, 2005; Jukes & Martinsen, 2007; Vemulapalli et al., 2017; da Graça Pereira et al., 2017; Zemanova et al., 2021; Woolcock & Lazarova, 2022).

The ethics of animal use in research and education are succinctly expressed by Russell and Burch's 3Rs rationale (Replacement, Reduction, Refinement) (Russell & Burch, 1959). These principles encourage, when possible, to replace the use of animals with non-animal alternatives, to reduce the number of animals to the minimum considered necessary, and to limit the pain and distress to which animals are exposed. Some researchers have suggested an additional "R" in animal experimentation, which stands for Responsibility, encompassing both ethical treatment of experimental animals and the maintenance of their social and scientific standing (Tannenbaum & Bennett, 2015). As a result, animal ethics committees typically assess research projects based on the principles of the 4 Rs (Lee et al., 2020). Accordingly, this approach articulates a particular ethical stance, providing deontological moral justifications to endorse the utilization of animals, in line with the principles of bioethics (Baldelli et al., 2019). To foster the development of ethical competence regarding the use of animals, which is a fundamental skill for students in both veterinary and humane medicine undergraduate programs, ethical topics related to the use of animals should be incorporated into their teaching curriculum (e.g. Lord et al., 2010; Pollard-Williams et al., 2014; Shivley et al., 2016; Baldelli et al., 2019).

In Portugal, the breeding, supply and use of animals for scientific purposes, namely research and education, is regulated by EU Directive 2010/63/EU of the European Parliament and of the Council of 22 September 2010, which was transposed to national law through Decree-Law No. 113/2013. The latter has already been amended by Decree-Law 1/2019, making adjustments to its articles, so that EU Directive 2010/63/EU is correctly interpreted and implemented. This legislation was created with the aim of improving and ensuring the welfare

of animals used for scientific purposes, promoting the concept of the Three Rs (Replacement, Reduction, Refinement) and their humane, reflected, responsible and justified use. Decree-Law 113/2013 gives effect to the previously stated principles, namely with regard to the choice of procedures to be applied, giving preference to the use of alternative methods.

Several studies have assessed the attitudes of students, and / or other members of the academic community, about the use of animals in higher education for educational purposes and / or scientific experimentation (eg. Paul & Podberscek, 2000; Donaldson & Downie, 2007; Knight et al. 2010; Hazel et al., 2011; Phillips et al., 2012; Pollard-Williams et al., 2014; Lairmore & Ilkiw, 2015; Minor-Campos et al., 2019a, b; Andreoli et al., 2023), but questionnaires focusing on the perspective of veterinary and medical students regarding the use of higher education institutional animals, bred and kept in captivity specifically for educational purposes (e.g. kennels at the Universities), are not known.

The objective of this research was to characterize the utilization of animals (yes/no, species used, procedures performed) in the undergraduate training programs of the Integrated Master Degrees in Veterinary Medicine (MIMV) students from the Abel Salazar Institute of Biomedical Sciences (ICBAS) - University of Porto and from the University of Trás-os-Montes and Alto Douro (UTAD), as well as the Integrated Master Degrees in Medicine (MIM) students from ICBAS and from the Faculty of Medicine of the University of Porto (FMUP). Additionally, we intended to document the ethical, pedagogical, and animal welfare perspectives of veterinary and medical undergraduate students concerning the use of animals exclusively for educational purposes (excluding the clinical context in the case of MIMV). It is estimated that this knowledge may help teachers to gain an insight of the student's sensitivities, raising awareness within the academic community about ethical and justified animal use, and ultimately promoting the implementation of non-animal educational strategies in the veterinary and human medicine undergraduate educational programs.

Material and Methods

With the objective of understanding the perspective of undergraduate students of Medicine and Veterinary Medicine, and characterizing their studies cycle regarding the use of animals for an exclusively educational purpose, a survey was developed by the authors, between September 2018 and April 2019, with open and closed questions (Likert scale 1–5 and yes / no). For its optimization, in May 2019 the questionnaire was disseminated on paper to 16 ICBAS undergraduate veterinary volunteer students, due to ease of access and convenience. The form was completed in person, so that the students could express any doubts regarding the interpretation of the questions, also allowing to determine the average time to fill it. This process conducted to some minor semantics improvements to the questionnaire.

In June/July 2019, after approval by the Ethics Committee of the São João Hospital Center / Faculty of Medicine of the University of Porto (n° 120/19), the survey was disseminated on paper to 100 undergraduate MIM students of ICBAS (n = 40) and FMUP (n = 60) and 80 undergraduate MIMV students of ICBAS (n = 40) and UTAD (n = 40). To the MIM ICBAS students, the survey was delivered after a practical class, where animals have been used with pedagogical purposes, and optionally returned till the end of the same week. In every other case, questionnaires were immediately returned after completion. Students were informed ("Explanatory notes", first page of the document) that the survey was being conducted for a master's degree thesis and that they could withdraw from the study at any time without causing any harm. All questionnaires were

anonymous and additional comments were optional (last page of the document). This study analyzed the majority of the questions surveyed, but not all, in order to accurately address the goals of the present research.

Statistical analysis of the survey was performed (IBM SPSS version 26 program) in order to identify response trends and clarify the perspective of MIM and MIMV students regarding the use of animals bred and kept captive uniquely for educational purposes. The descriptive statistics of the sample allowed the characterization by:

1. The gender, age, number of students in each institution and their study cycle (Question 1);
2. The level of agreement with the use of animals (Likert scale: totally agree, agree, no opinion, disagree, totally disagree; yes/no: if there is no alternative), and which animals (yes/no: all; rodents, batrachians and fishes, horses, cows, pigs, sheep and goats, dogs and cats, non-human primates, birds and rabbits), exclusively for educational purposes (Questions 2 and 2.1);
3. Types of animals used in classes and types of procedures performed (open questions) (Questions 3 and 4);
4. The usefulness of including animals in class (Likert scale: always, very often, no opinion, few times, never) (Question 5);
5. Students' knowledge of possible alternatives to animal use and willingness to implement them (open questions) (Questions 6 and 7).

For questions regarding the degree of agreement on the use of animals for educational purposes, their usefulness and alternative methods (questions 2, 5, 6 and 7), a comparison was made between groups according to academic background (MIM or MIMV), institutions of origin (UTAD, ICBAS, FMUP), and gender, using the t-student test, treating the Likert variables as continuous, and the chi-square test (χ^2) treating these variables as categorical. In situations where the expected frequencies were lower than 5, Fisher's exact test was used. Comparison between more than two groups was performed using the nonparametric Kruskal-Wallis and Mann-Whitney tests. Data is presented in figures and tables with the numbers and percentages of respondents in each category. The significance level considered was 0.05.

We also transcribed and analyzed some opinions of the students who, in the author's opinion, translate different perspectives or include key points about the discussion of this theme, namely regarding:

1. The inclusion of clinical practice in the hospital (clinical context) or outpatient setting as an alternative method to the use of animals exclusively for educational purposes (in case of MIMV students);
2. The opinions in agreement with the use of animals;
3. The opinions in disagreement with the use of animals;
4. The overall opinions about the use of animals as a teaching methodology.

Legally, "procedure is any use, invasive or not, of an animal for experimental or other scientific purposes, with known or unknown results, or for educational purposes, which may cause a level of pain, suffering, distress or lasting damage equivalent to or greater than that caused by the introduction of a needle in accordance with good veterinary practices,

including any action intended or likely to lead to the birth or hatching of an animal, or the creation and maintenance of a genetically modified animal lineage, excluding the slaughter of animals solely for the use of their organs or tissues” (Decree-Law 113/2013). According to article 15 of Decree-Law 113/2013, each procedure will be classified according to the severity that can be caused to an animal, according to the following categories:

1. Non-recovery: procedures performed entirely under general anesthesia, from which the animal does not regain consciousness;
2. Mild: procedures performed on animals that are likely to cause them to experience mild pain, suffering or short-term distress, as well as procedures without significant harm to the welfare or general condition of the animals;
3. Moderate: procedures that are performed on animals and are likely to cause them to experience moderate short-term pain, suffering or distress, or mild long-term pain, suffering or distress, as well as procedures likely to cause moderate welfare damage or general condition of the animals;
4. Severe: procedures performed on animals that are likely to cause them severe pain, suffering or distress, or moderate long-term pain, suffering or distress, as well as procedures likely to cause severe damage to welfare or general condition of the animals.

For the purpose of systematization and simplification of the description of the procedures performed on animals, they were grouped into the following three categories (Question 4):

1. Live animals: included procedures such as noninvasive physical examination, rectal palpation, vaginal cytology, skin scraping, ultrasound, radiography, anesthesia, surgery, oral or parenteral administrations such as intravenous (IV), subcutaneous (SC), or intramuscular (IM), others;
2. Use of the cadaver: animals sacrificed specifically for the class;
3. Sacrificed animals: animals euthanized after performing procedures while alive.

Results

Most questionnaires (139 out of 180) were returned partially or fully completed, with the number of respondents varying for each question. For MIMV and MIM at ICBAS and MIMV at UTAD, 40 surveys were disseminated for each study cycle, resulting in response rates of 90%, 35% and 90%, respectively. In the case of MIM at FMUP, an 88.3% response rate was achieved after the distribution of 60 questionnaires.

Sample Characterization (Question 1)

The study sample included 26 male and 113 female students (81.29%, 113/139), with a median age of 22.61 years-old (range 20–35 years-old). Sixty students, all from MIM (n=48 from FMUP and n=12 from ICBAS), were attending the 3rd year of the study cycle, 24 the 4th year (n=5 MIM FMUP, n=2 MIM ICBAS and n=17 MIMV ICBAS) and 55 the 5th year (n=36 MIMV UTAD and n=19 MIMV ICBAS).

Level of Agreement with the Use of Animals, and which Animals, Exclusively for Educational Purposes (Question 2)

Most students ($n = 86/131$, 65.65%) agreed to the establishment, maintenance and performance of animal procedures solely for educational purposes as a way of guaranteeing optimal acquisition of theoretical knowledge, attitudes and behaviors and technical skills (Table 1).

The significance of responses to this question was analyzed comparing groups by study cycle, institution and gender. There are no statistically significant differences between genders ($\chi^2 = 3.742$, $p = 0.442$) but there are statistically significant differences between study cycles/institutions (Kruskal-Wallis test: $\chi^2 = 9.511$, $p = 0.023$). MIMV UTAD students agreed more with the use of animals for educational purposes than students from other institutions (88.6% agreed or completely agreed). On the other hand, it was evidenced that medical students have more dispersed opinions regarding this use compared to veterinary medicine students, 78.2% of whom agreed or completely agreed (statistically significant difference: $\chi^2 = 18.967$, $p = 0.001$; Mann-Whitney Z test = -2.074 , $p = 0.038$). Thus, veterinary medicine students presented the highest agreement regarding the use of animals for educational purposes compared to those of medicine and, of the former, UTAD students were those who had a more favorable opinion of this use (Table 1).

Most students (118/139; 84.9%) agreed with the use of animals exclusively for educational purposes if there are no alternative methods that can substitute them, but 64% only agreed with the use of some species ($n = 89/139$); rodents, batrachians and fishes were the most validated, whereas non-human primates, dogs and cats were the most protected species (Fig. 1).

In these questions we have identified some contradictory answers. For instance, 27 students ($n = 27/139$, 19.4%) agreed with the use of all animals; of these, 25/27 actually answered “yes” for all the indicated species, but 2 answered “no”; one in dogs and cats ($n = 1/27$) and another in non-human primates ($n = 1/27$). The remaining 112/139 agree with the use of some species ($n = 89/112$) or none ($n = 23/112$). Again, 11/112 students, who indicated that they did not agree with the use of all species, selected “yes” in all species listed in the survey ($n = 2$ MIMV ICBAS, $n = 6$ MIMV UTAD and $n = 3$ MIM FMUP).

Table 1 Distribution of the frequency(percentage) of answers to question 2, according to the study cycle/institution: “Do you agree with the establishment, maintenance and performance of procedures on animals exclusively for educational purposes?”

SCycle / Institution	TD	D	NO	A	TA
MIMV ICBAS, n = 34	1(2.9)	10(29.4)	0	18(52.9)	5(14.7)
MIMV UTAD, n = 35	0	4(11.4)	0	21(60.0)	0(28.6)
MIM ICBAS, n = 14	1(7.1)	5(35.7)	2(14.3)	4(28.6)	2(14.3)
MIM FMUP, n = 48	3(6.3)	10(20.8)	9(18.8)	16(33.3)	10(20.8)
TOTAL, n = 131	5(3.8)	29(22.1)	11(8.4)	59(45.0)	27(20.6)

SCycle study cycle, TD, I totally disagree, D I disagree, NO No opinion, A I agree, TA I totally agree, MIM Integrated Master in Medicine, MIMV Integrated Master in Veterinary Medicine, ICBAS Abel Salazar Institute of Biomedical Sciences, UTAD University of Trás-os-Montes and Alto Douro, FMUP Faculty of Medicine of the University of Porto

Question 2.1

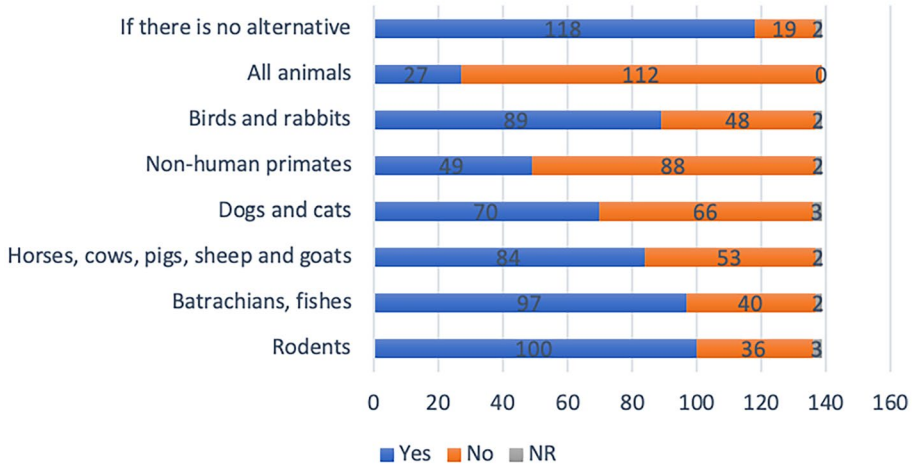


Fig. 1 Number of responses to question 2.1. concerning the agreement (yes/no) with the use of animals for educational purposes according to the different species and the existence, or not, of alternative teaching methods; NR: Non responder

Types of Animals Used in Class and Types of Procedures Performed (Questions 3 and 4)

MIM and MIMV students (n=79/139, 56,8%) reported procedures performed with a pedagogical purpose in animals such as rodents, dogs, cats, birds, rabbits, cows, pigs, goats, horses, drosophilas, fishes, wild animals, batrachians, and sheep (Tables 2, 3, 4 and 5; Fig. 2).

Notwithstanding what is described in Table 5, the majority (n=41/53, 73.36%) of the students refer that in MIM FMUP base curriculum there are no contents in which animals are utilized for teaching purposes, in the context covered by this survey. However, they have mentioned that there are optional curricular units in which animals are used, but they are scarce (none was specified in the questionnaires received).

All procedures were categorized of “not applicable” nature to the legislative context, of “non-recovery” or of a “mild” to “moderate” degree of severity (Tables 2 to 5 and Fig. 2). However, it was not always possible to group the type of procedures performed, since some students do not describe the actual use, just referring if the animal was alive, if they

Table 2 Procedures performed in curricular units that use animals for educational purposes in MIM ICBAS and corresponding classification of the degree of severity in accordance with Decree-Law No. 113 of 2013 (determined when possible)

Animals	Procedures described by MIM ICBAS students
Rodents	Animal sacrificed after performing the procedure while alive (NR); Animal sacrificed for class (NA), after use in a scientific research project
Batrachians	Animal sacrificed for class (NA)

NA Not applicable, that is, it is not a “procedure” as defined in the legislation, NR non-recovery

Table 3 Procedures performed in curricular units that use animals for educational purposes in MIMV ICBAS and corresponding classification of the degree of severity in accordance with Decree-Law No. 113 of 2013 (determined when possible)

Animals	Procedures described by MIMV ICBAS students
Rodents, dogs	Non-invasive physical examination (NA), acupuncture (L), ECG (NA)
Batrachians	Animal sacrificed for class (NA); animal sacrificed after performing a procedure while alive (O ₂ consumption) (perhaps NR)
Rodents, sheep, goats, cows, horses, birds, dogs, rabbits	IV, IM, SC (L) administrations; animal sacrificed after performing a procedure while alive (perhaps NR); Animal sacrificed for class (NA), after use in a scientific research project
Sheep, cows, horses	Anesthesia (L)
Cows, horses	Surgery (L or M#), anesthesia (L), drug administration (L)
Dogs, cats	Non-invasive physical examination (NA), skin scraping (L), rectal palpation (NA or L*)
Dogs, horses	Ultrasound (NA), X-rays (NA+)
Cows, horses	Non-invasive physical examination (NA), rectal palpation (NA or L*)
Cows, horses	Surgeries (L or M#)
Dogs, cats	Non-invasive physical examination (NA), ultrasound (NA), radiography (NA+), drug administration (L)
Dogs, cats	Anesthesia (L), surgery (L or M#)
Dogs, cows and horses	Rectal palpation (NA or L*), vaginal cytology (NA), ultrasound (NA), semen collection (NA)

NA Not applicable, that is, it is not a “procedure” as defined in the legislation; +: Not applicable if sedation/anesthesia has not been performed (in the latter case, it would be “L”); NR non-recovery; L Mild; *: may be mild depending on user’s experience and number of students for the same animal (author’s note); otherwise, it will be NA; #: depends on the type of surgery; M Moderate; S Severe. ECG Electrocardiogram

used the cadaver or both. On the other hand, unavailable information, such as the duration and repetition of each procedure may contribute to increase its severity, regardless of the manipulation itself.

Usefulness of Including Animals in Class (Question 5)

A total of 65.5% of all respondents (n = 72/110) considered that the use of animals was valuable for their learning outcomes (Table 6). The relevance of the responses to this question was also analyzed by comparing the groups by study cycle, institution and gender. There are no statistically significant results between genders ($\chi^2 = 7.223$, $p = 0.125$; Fisher’s exact test 7.205, $p = 0.100$) but there are statistically significant differences between study cycles/institutions (Kruskal-Wallis test, $\chi^2 = 16.732$, $p = 0.001$), namely amongst:

1. MIMV ICBAS and MIMV UTAD ($\chi^2 = -14.869$, $p = 0.041$); UTAD students consider that learning with animals is more useful than ICBAS students;
2. MIMV ICBAS and MIM FMUP ($\chi^2 = 15.346$, $p = 0.041$); this difference is due to the fact that FMUP students show a high percentage of “no opinion” answers versus ICBAS students who, mostly (73.5%), find a great use in learning with animals;
3. Medical students and Veterinary Medicine students (Fisher’s exact test 29.690, $p < 0.001$); the latter found a greater utility in learning with animals than medical students.

Table 4 Procedures performed in curricular units that use animals for educational purposes in MIMV UTAD and corresponding classification of the degree of severity in accordance with Decree-Law No. 113 of 2013 (determined when possible)

Animals	Procedures described by MIMV UTAD students
Dogs, cats, horses, cows, sheep, goats	Use of cadaver, animal sacrificed for class (NA)
Fish, rodents	Assessment of physiological parameters (eg temperature measurement) changing environmental conditions (NA), anesthesia (L)
Horses, cows	Procedures with the live animal: observation of the external morphology (NA)
Dogs, horses, cows	Non-invasive physical examination (NA), ultrasound (NA)
<i>Drosophila</i> (insect)	Animal sacrificed after performing procedures while alive (??)
Dogs, birds, pigs, cows, horses	Live animal procedures (??)
Dogs, cats	Live animal procedures (??)
Dogs, horses	Ultrasound (NA)
Dogs, cats, sheep, birds, rabbits, wild animals	Use of the cadaver after natural death or euthanasia (NA); use of cadaver, animal sacrificed specifically for class (NA)
Dogs, horses, cows	Physical examination (NA), oral administrations (NA), simulated drug administration (L)
Dog, cat, horse, cow, sheep, goats	Physical examination (NA), skin scraping (L)
Dog, cat, horse, cow, sheep, goats	Physical exam (NA)
Dogs, cats	Use of cadaver after natural death or euthanasia (NA), anesthesia (L), surgeries (L or M#)
Dogs	Physical examination (NA), drug administration (L), skin scraping (L)
Horses	Physical examination (NA), radiographs (NA), ultrasound (NA), dressings (NA), anesthesia (L)
Cows	Hoof trimming (NA)
Cows	Rectal palpation (NA or L*)
Dogs, cows, horses, sheep, goats	Ultrasound (NA), rectal palpation in cows (NA or L*), vaginal cytology (NA), physical examination (NA)
Dogs, cats	Physical examination (NA), cytology (L), skin scraping (L), ultrasound (NA), computed tomography (L), radiographs (NA+), ECG (NA)
Fishes	Use of cadavers, animal sacrificed specifically for class (NA); animal sacrificed after performing procedures while alive (??)
Cows, horses, pigs, sheep, goats, chickens, fish	Fish dissection (NA), live animal procedures (??); animal sacrificed after performing procedures while alive (??)

NA Not applicable, that is, it is not a "procedure" as defined in the legislation; +: Not applicable if sedation/ anesthesia has not been performed (in the latter case, it would be "L"); NR non-recovery, L: Mild; *: may be mild depending on user's experience and number of students for the same animal (author's note); otherwise it will be NA;?: examples of procedures performed are unknown; #: depends on the type of surgery; M: Moderate; S: Severe. **The UTAD MIMV study cycle includes four internships (2nd, 4th and 5th year). ECG Electrocardiogram

Table 5 Procedures performed in curricular units that use animals for educational purposes in MIM FMUP and corresponding classification of the degree of severity in accordance with Decree-Law No. 113 of 2013 (determined when possible)

Animals	Procedures described by MIM FMUP students
Rodents, birds (chicks)	Use of cadavers, animal sacrificed specifically for class (NA); animal included in a research study, so it was not sacrificed specifically for the class (NA); genetic studies
Rodents, rabbits	Use of cadavers, animal sacrificed specifically for class (NA)
Rodents, rabbits	Use of cadaver, animal sacrificed specifically for class (NA); histological preparations, do not know tissue origin

NA: Not applicable, that is, it is not a “procedure” as defined in the legislation

Students’ Knowledge of Possible Alternatives to Animal Use and Willingness to Implement them (Questions 6 and 7)

The majority ($n=73/126$, 57.9%) of students did not recognize alternative methods that could replace the use of live animals, or animals sacrificed for teaching purposes, in the classes they attended. Those 53 who answered “yes” mentioned videos of invasive

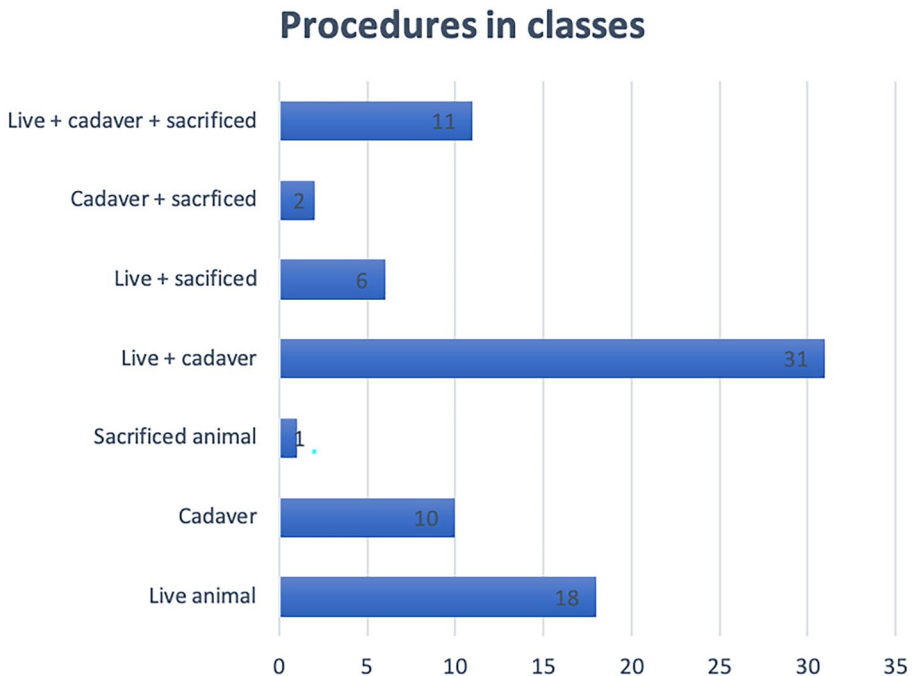


Fig. 2 Procedures performed in classes grouped by categories; with the “live animal”: non-invasive physical examination, rectal palpation, vaginal cytology, skin scraping, ultrasound, radiography, anesthesia, surgery, drug administrations (IV, SC, IM, oral), others; use of the “cadaver”, an animal sacrificed specifically for the class; animal “sacrificed” after performing procedures while alive

Table 6 Distribution of the frequency(percentage) of responses to question 5 ("Did you consider the use of animals useful for your learning outcomes?"), according to each study cycle/institution

SCycle / Institution	Never	Few times	NO	Very often	Always
MIMV ICBAS, n = 34	0	5(14.7)	4(11.8)	20(58.8)	5(14.7)
MIMV UTAD, n = 35	0	4(11.4)	0	17(48.6)	14(40.0)
MIM ICBAS, n = 10	0	3(30.0)	1(10.0)	4(40.0)	2(20.0)
MIM FMUP, n = 31	3(9.7)	1(3.2)	17(54.8)	6(19.4)	4(12.9)
TOTAL, n = 110	3(2.7)	13(11.8)	22(20.0)	47(42.7)	25(22.7)

NO No opinion, *MIM* Integrated Master in Medicine, *MIMV* Integrated Master in Veterinary Medicine, *ICBAS* Abel Salazar Institute of Biomedical Sciences, *UTAD* University of Trás-os-Montes and Alto Douro, *FMUP* Faculty of Medicine of the University of Porto

procedures (n = 21), three-dimension anatomic models (n = 7), mannequins (n = 6), models specifically for drug administration (n = 6), rectal palpation simulator (n = 5), clinical context in case of MIMV (n = 4; training of clinical procedures such as venipuncture, sutures, physical exam, or others, in clinical environment), dissection of animals ethically obtained (n = 4; not sacrificed for the purpose: euthanasia or natural death, research animals), theoretical knowledge per se could be enough for learning outcomes (n = 4).

The significance of the responses regarding knowledge of the existence of alternative methods to the use of animals was analyzed by comparing the study cycle, institution and gender groups. Statistically significant results concerning genders ($\chi^2 = 0.099$, $p = 0.753$) were not observed, but statistically significant differences between study cycles/institutions were identified ($\chi^2 = 40.751$, $p < 0.001$), namely MIMV students were more aware about alternative methods to animal use than MIM students ($\chi^2 = 18.855$, $p < 0.001$); MIMV ICBAS students have a higher awareness about alternative methods than MIMV UTAD students ($\chi^2 = 14.619$, $p < 0.001$), and MIM FMUP students have the minor knowledge about alternative methods (Table 7).

Students were also questioned about the option of using alternative methods and their possible impact on the acquisition of theoretical knowledge, attitudes and behaviors and technical skills (question 7). Most respondents (n = 92/137, 67.15%) found that alternative methods should be used if the learning process was not compromised (15.33% consider that they should always be used). Statistically significant results were found between

Table 7 Distribution of the frequency(percentage) of answers to question 6, ("Do you know alternative methods that could replace the use of live animals, or animals specifically sacrificed for pedagogical purposes, in the classes you attended?"), according to the study cycle/Institution

SCycle/Institution	No	Yes
MIMV ICBAS, n = 34	6(17.6)	28(82.4)
MIMV UTAD, n = 35	22(62.9)	13(37.1)
MIM ICBAS, n = 13	6(46.2)	7(53.8)
MIM FMUP, n = 44	39(88.6)	5(11.4)
TOTAL, n = 126	73(57.9)	53(42.1)

MIM Integrated Master in Medicine, *MIMV* Integrated Master in Veterinary Medicine, *ICBAS* Abel Salazar Institute of Biomedical Sciences, *UTAD* University of Trás-os-Montes and Alto Douro, *FMUP* Faculty of Medicine of the University of Porto

Table 8 Frequency(percentage) of answers, according to gender, regarding the possibility of animals being substituted as teaching methodologies

Gender	Substituted without prejudice	Substituted with prejudice	Cannot be substituted	I don't know
Male, n = 23	12(46.2)	3(11.5)	6(23.1)	5(19.2)
Female, n = 103	80(72.1)	18(16.2)	5(4.5)	8(7.2)
TOTAL, n = 126	92(67.2)	21(15.3)	11(8.0)	13(9.5)

genders ($\chi^2 = 14,667$, $p = 0,002$; Fisher's exact test 12,713 and $p = 0,003$) because there was a higher difference in the number of answers given by female students for each of the options, and the distribution of responses from male students was more homogeneous. Nevertheless, in both genders predominated the preference for the option "animals can be substituted if learning is not compromised" (females 72.1%; males 46.2%) (Table 8).

In question 7, statistically significant results were also observed between the different study cycles/institutions ($\chi^2 = 18.062$, $p = 0.034$; Fisher's exact test 15.930, $p = 0.043$) (Table 9), namely between medical and veterinary medicine undergraduate students ($\chi^2 = 8.981$, $p = 0.03$).

Student's Comments

Student's comments that translate different perspectives or include key points about the discussion of the use of animals in pedagogical context are presented in Table 10. Information was grouped by study cycle (MIM and MIMV) in four categories, namely:

1. Clinical context presented as an alternative method;
2. Opinions in agreement with the use of animals;
3. Opinions in disagreement with the use of animals;
4. Overall opinions about using animals in pedagogical context.

Table 9 Frequency(percentage) of responses, according to the study cycle/institution, regarding the possibility of animals being substituted as teaching methodologies

SCycle/Institution	Substituted without prejudice	Substituted with prejudice	Cannot be substituted	I don't know
MIMV ICBAS, n = 34	25(73.5)	5(14.7)	2(5.9)	2(5.9)
MIMV UTAD, n = 36	27(75.0)	4(11.1)	5(13.9)	0
MIM ICBAS, n = 14	11(78.6)	3(21.4)	0	0
MIM FMUP, n = 53	29(54.7)	9(17.0)	4(7.5)	11(20.8)
TOTAL, n = 137	92(67.2)	21(15.3)	11(8.0)	13(9.5)

MIM Integrated Master in Medicine, MIMV Integrated Master in Veterinary Medicine, ICBAS Abel Salazar Institute of Biomedical Sciences, UTAD University of Trás-os-Montes and Alto Douro, FMUP Faculty of Medicine of the University of Porto

Table 10 Student's opinions/comments about the use of animals in a pedagogical setting

Subjects discussed	MIMV students	MIM students
Clinical context	<p>“...students should accompany veterinarians in clinical context as soon as possible to observe and train procedures”.</p> <p>“...traineeships with a longer period of attendance... at the Hospital...could allow for practical learning and the consolidation of knowledge in order to dispense the use of animals exclusively for educational purposes, such as, for example, to practice administrations.”</p> <p>“Animals must be substituted whenever there are alternatives, even if learning is a little compromised, which can be compensated later in clinical practice”.</p> <p>“The use of animals for educational purposes can be essential in some situations, but the ideal is to watch procedures in a clinical context and proceed, step by step and with the help of teachers and clinicians, improving both knowledge and technique”.</p>	<p>Not applicable.</p>
Agree with use	<p>“The use of animals in the context of the class, even if it causes some stress to the animal, will be useful to save a greater number of animals in the future”.</p> <p>“At the beginning of the training, it is justified to reduce the number of cadavers and live animals, but these are always necessary in a more advanced phase”.</p> <p>“There is nothing that can replace a live animal.”</p> <p>“Due to the nature of the veterinary medicine degree, the use of animals is very important (the acquisition of technical knowledge implies working with patients)”.</p> <p>“I consider that it is very difficult to replace learning procedures without dealing with animals. Otherwise, in our professional life we would not be prepared to perform them in sick animals”.</p>	<p>“Regardless of the ethics involved, I think it is important to learn the human constitution through models with similar characteristics, but if there is another way to continue doing this without using animals, I would support it”.</p> <p>“Sometimes there is no other alternative method and it is important, above all, to obtain knowledge of the best possible quality”.</p> <p>“Learning in living models allows the acquisition of other types of skills than non-living models”.</p> <p>“There are certain experiments that require the effect on live animals”.</p> <p>“Despite caring for animals rights, I think they are a fundamental instrument of learning”.</p>

Table 10 (continued)

Subjects discussed	MIMV students	MIM students
Disagree with use	<p>“It is essential not to compromise animal welfare, even if it serves educational purposes”.</p> <p>“Nowadays, animal sacrifice is not justified because certain experiences can be simulated by watching videos”.</p>	<p>“I do not agree with its use, theoretical knowledge is enough”.</p> <p>“I believe that learning should not depend on the use and disrespect of other forms of life”.</p> <p>“Animals are often subjected to mistreatment and sub-optimal conditions, which does not justify the gain in terms of learning, taking into account that there are alternative models and methods”.</p> <p>“The suffering of living beings must be avoided as much as possible, even at the expense of education”.</p>
Overall opinions	<p>“...learning is very dependent on practice; therefore, models should only be used if there is no impairment of learning...”.</p> <p>“Surgery and administration models are desirable to reduce training in live animals...”.</p> <p>“I don't imagine if there is any model that can replace everything that is learned using live animals, however some things I agree that can be replaced”.</p> <p>“...dealing directly with animals promotes a greater motivation and learning...”.</p> <p>“The use of models may interfere with the commitment to learn since the student is aware that he is not manipulating an animal, therefore, he is not responsible for the procedure or does not perform it with proper care and attention”.</p> <p>“It is a controversial subject and its solution will not necessarily be black or white. Each situation must be analyzed from the different perspectives”.</p> <p>“...teachers should accompany students individually to help them to improve contact and procedures with animals...”.</p>	<p>“Animals have as many rights as we do”.</p> <p>“Avoiding animal sacrifice without harming knowledge is beneficial”.</p> <p>“There are situations in which learning can only be theoretical, with no need to sacrifice animals”.</p> <p>“Despite considering that the use of animals was very useful for our learning and, in part, agreeing with their use for scientific purposes, it is crucial to recognize that animals should be replaced by other methods. In terms of scientific research, in my view there is no possible replacement. However, in education there is no need to repeatedly sacrifice animals every year”.</p> <p>“Until the time of my training, I had never worked with animal models in experimental laboratory work”.</p> <p>“Students must be taught with the best existing method so they can be the best professionals”.</p>

Discussion

For decades, many thinkers and philosophers (Singer, 1975; Regan, 1983; Carruthers, 1992; Scruton, 2000; Fox, 2002) have proposed ethical profiles/views to characterize citizens' perspectives on sensuous animal experimentation, which are summarized and described in the Animal Ethics Dilemma interactive learning tool for university and professional training (website Animal Ethics Dilemma, 2023):

Contractarian view, morality is based on agreements/contracts with mutual benefits (e.g.

“To improve the quality of animal research, one should be concerned about animal welfare.”).

Utilitarian view, morality is based on optimizing human and animal well-being (e.g. “Some animal research may be justified by its vital importance, as it may enable us to find cures for alleviate painful diseases.”).

Relational view, morality is based on the degree of relationship between animals and humans, and humans to each other (e.g. “A dog is a man’s best friend, so it should be treated better than animals on farms and in laboratories.”).

Animal rights (abolitionist) view, morality cannot be justified by good results if it implies resorting to cruel means (e.g. “There is no moral justification for any experimentation detrimental to animals, excluding for their own benefit”).

Respect for nature view, morality is based on the belief that we must respect not only individual animals but also their species (e.g. “Nature must take its course.”).

In this study, the majority of the students (65.65%) revealed a predominant utilitarian ethical profile, meaning that they consider that animal use in pedagogical context is ethically acceptable if the benefits outweigh the costs, provided that refinement of animal procedures is ensured (utilitarian cost-benefit analysis). However, for each pedagogical procedure using animals (excluding the clinical context, in case of MIMV students) a major challenge may be to objectively characterize the “benefit” (learning outcomes) versus the “cost” (harms to the animals utilized). To establish the former, comparative studies (animal-based versus alternative training methods) need to be conducted in order to determine that animal use cannot be substituted (3Rs policy “Replacement”). To address the “cost”, 3Rs “Reduction” and “Refinement” must also be considered (Knight, 2011). Accordingly, we may question if, in the pedagogical context, the use of animals is humane, rational and justified? Do learning outcomes outweigh the respect for life?

Although most of the students agree with the use of animals for educational purposes, 64% (n = 89/139) only agree with the use of some species, also evidencing a relational ethical profile (most protected animals were non-human primates, dogs and cats; conversely, rodents, batrachians and fish were the most legitimated). By analyzing the data published by the Portuguese General Directorate for Food and Veterinary (DGAV) (website DGAV, 2023), rodents are the most used species in pre- and post-graduate education, so it is possible that students have difficulty in acquiring and developing balanced attitudes towards animal experimentation, if there is a regular use of animals of a particular species (Van der Valk et al., 1999). Nevertheless, is it legitimate to say that the life and well-being of a dog is more important than that of a mouse?

In the present context, alternatives are methodologies or teaching approaches that replace the harmful use of animals, by promoting a more humane education in life sciences, without compromising the objectives of teaching (Russell & Burch 1959; Van der

Valk et al., 1999; website ECVAM, 2023). For some authors, these progressive educational methodologies should give freedom of conscience to students, encouraging a holistic perception and respect for life (Jukes & Chiuria, 2003; Woolcock & Lazarova, 2022). A high percentage ($n=73/126$, 57.9%) of respondents of this study are not aware of alternative methods that could replace the use of live animals, or animals sacrificed for this purpose, in the classes they attended. This could be justified by the fact that the majority of FMUP students did not answer this question, since they did not use animals in their pre-graduation training. On the other hand, MIMV ICBAS students demonstrated a more consolidated knowledge about the existence of alternative methods compared to those from UTAD. This statistically significant difference seems to show a greater awareness of the MIMV ICBAS students to the subject, most likely because they were exposed to such themes through curricular or extracurricular activities and events, as well as may explain the greater utility in the use of live animals reported by the MIMV UTAD students. Nevertheless, we believe that this consciousness will progressively increase since EU Directive 2010/63/EU and Decree-Law n° 113/2013 clearly state that “animals have an intrinsic value, which must be respected, and their use in procedures raises ethical concerns, so they must be treated as sentient creatures. Their use in procedures should be limited to contexts where such use provides benefits to human, animal health or the environment. Consequently, the use of animals for scientific or educational purposes should only be considered when a non-animal alternative does not exist”. In accordance, several EU governments may be committed to promote and implement alternative methods in both research and education (e.g. Andreoli et al., 2023). In Portugal, Animal Welfare Bodies (ORBEA) can and should have an increased responsibility in the dissemination and implementation of alternative pedagogical methodologies to the use of animals in the respective Institution (Decree-Law, 113/2013; Dispatch, 2880/2015). Nevertheless, teachers also have a moral and ethical responsibility to promote conscientious pedagogical practices with (or without) animals, contributing to the education of the following generations of veterinary and medical students on essential ethical values, namely respect for life (Baptista, 2019).

The European Centre for the Validation of Alternative Methods (ECVAM) (website ECVAM, retrieved 15 June 2023) has as main objective to promote and regulate the scientific acceptance of alternative methods that are important for the biosciences and that reduce, refine or replace the use of laboratory animals. One of the first priorities defined by ECVAM was the implementation of procedures that would allow obtaining grounded information about the state of the art in the development and validation of non-animal tests and their potential for their eventual incorporation in routine procedures. Thus, multiple alternative resources to the use of animals have been identified, with their applicability and effectiveness being progressively tested in an educational context, namely models, mannequins and mechanical simulators; interactive movies and videos; computer simulations and virtual reality systems; self-experimentation and human studies; experiments with plants; observational and field studies; systems biology; animal sub-products from slaughterhouses and fishing docks; in vitro studies in cell lines; ethically obtained cadavers; molecular tools, functional genomics; tissue engineering; clinical practice, among others.

The alternative method most cited by students was viewing explanatory videos of pertinent laboratory or clinical procedures ($n=21/139$, 15.11%), but models, mannequins, simulators, cell cultures, cadavers and learning in clinical context were also listed. Four students mentioned that, under certain circumstances, theoretical exposition may be sufficient for learning, through reading and analyzing scientific articles or illustrative schemes. Indeed, videos allow to document experimental or clinical procedures, reducing the number of animals or even replacing them (Fawver et al., 1990). Models are used to study anatomy and perform

procedures without the stress that comes with this type of manipulation. For example, three-dimensional models of plastinated animals are available (plastination is a chemical process that transforms the tissues of a dead animal into plastic) or mannequins (anatomical models), such as a life-size dog designed for training of cardiopulmonary resuscitation, or models for dissection or intubation of neonates (Haspel et al., 2013; Gayson et al., 2015). In turn, simulators provide a potential solution to some of the challenges in teaching anatomy to students of Medicine or Veterinary Medicine. Thus, several virtual reality simulators have been described in multiple contexts, namely, for performing surgeries, teaching the anatomy of the bovine abdomen (Kinnison et al., 2009) or for rectal palpation in cattle (Baillie et al., 2010), among others (Samsel et al., 1994; Cross & Cross, 2004; Behrend & Rosenthal, 2007), being that its use may not require the presence of instructors, therefore, it allows greater availability for training students, probably representing a more sustainable option for institutions (e.g. Baillie et al., 2010). Indeed, one of the most important arguments against the implementation of non-animal alternatives is their high cost, as well their validity as pedagogical tool. In fact, in a recent systematic review about the educational efficacy of non-animal teaching methods (Zemanova & Knight, 2021), in 90% of studies, humane methods such computer simulations, models, or videos were either equally effective or even more effective in achieving learning outcomes compared to harmful animal use. Conversely, only 10% of studies ($n=5$) in the field of anatomy, physiology, and veterinary surgical training presented higher efficacy of harmful animal use, which can be explained by inappropriate application or inadequate design of the non-animal teaching method (Zemanova & Knight, 2021). As evidenced, alternative humane methods may be suitable for certain types of educational learning objectives but not for others, having this selection a paramount importance in this discussion. Although sometimes challenging, the validity and reliability of these educational methodologies must be established, as preconized by ECVAM.

The cadavers used must be humanely and ethically sacrificed animals (e.g. animals that died naturally or that were euthanized following responsible scientific procedures). The importance of ethically sourced cadavers and their implications for teaching and education are exhaustively described by Martinsen and Jukes (2007), but generally, cadavers are used as alternative educational tools in veterinary schools to facilitate learning of animal anatomy and train surgical skills (Tefera, 2011; Woolcock & Lazarova, 2022). However, can an intentionally sacrificed animal, for pedagogical purposes, be categorized as ethically sourced? The fact is that the legislation in force (Decree-Law 113/2013) does not consider "the occision of animals solely for the use of their organs or tissues" as a "procedure", therefore, this use is not necessarily liable to be communicated or appreciated by the competent institutions, namely Animal Welfare Bodies (ORBEA) and the Portuguese General Directorate for Food and Veterinary (DGAV).

In the context of Veterinary Medicine, it was interesting to note that only 5.4% of the students surveyed mentioned "clinical practice" as an alternative method for teaching and learning subjects that necessarily involve handling and contact with animals. In fact, perhaps it was not properly apprehended that the carried-out survey only refers to animals used exclusively for educational purposes (clinical and other contexts are excluded), as explained in the instructions of the survey. Thus, by analyzing certain comments/observations, it is evident that some MIMV students considered this practice to be essential and, consequently, believed that the use of animals is indispensable for their veterinary training (showing a statistically significant difference when compared to their responses from medical students). Indeed, clinical practice is crucial to their professional education, and although alternative methods allow students to acquire fundamental skills and confidence in their execution, they are generally not adequate to completely replace the live animals

used to teach complex and multifaceted skills, such as management or surgical procedures (Da Graça Pereira et al., 2017). On the other hand, and as mentioned by one MIMV UTAD student, working with animals can promote understanding and retention of knowledge (Robinson et al., 2004; Ra'anan, 2005; Theoret et al., 2007; Shore et al., 2013), particularly those related to complex concepts and procedures (Ra'anan, 2005; Smeak, 2007; Drosdeck et al., 2013; Shore et al., 2013).

The (re)use of animals bred and kept in captivity for research, duly justified and approved by the ORBEA and/or DGAV (in case of Portugal), may contribute to reducing the number of animals sacrificed in classes (Van der Valk et al., 1999; Balcombe 2000). Thus, in this context, there are procedures performed on animals that are mild, so when the experimental protocol is concluded, these animals may be involved in classes with mild procedures or even in pedagogical activities that do not include “procedures” in the legal sense of the word. It is fundamental that the set of procedures or uses of animals be balanced and compatible with the best animal welfare practices, namely, with frequency and duration of utilizations suitably limited and adapted to each context. On the other hand, some experimental protocols include moderate to severe procedures and, as such, are potentially very debilitating for the animals. In the latter case, it is common for the animals to be sacrificed after the conclusion of the protocol, so they may, if appropriate, be subjected to some type of procedure in classes under general anesthesia and only subsequently euthanized (Baptista, 2019). Two MIM ICBAS students mentioned this type of use in rodents, in practical laboratory subjects of pharmacology. Additionally, as stated by four students, animals used in classes, which are kept alive after handling, should only be exposed to mild procedures, and any other use should be considered ethically reprehensible. In this research, all the procedures mentioned by the students were classified as mild, eventually moderate (in the case of some surgeries), and non-recovery, which may evidence a utilitarian use of animals for pedagogical purposes.

According to some of the students surveyed, it is understood as possible to replace the use of animals in the pre-graduate teaching of Veterinary Medicine and, certainly, in Medicine (as already occurs in MIM FMUP). Indeed, it would be wise to make a gradual transition from acquiring technical skills in non-animal methods, and then practicing in a clinical context, in the case of Veterinary Medicine students. Thus, in the first years of the undergraduate course, these students will be able to acquire clinical skills through the use of ethically obtained cadavers and/or their sub-products, synthetic models or computer simulators. Subsequently, it will be appropriate to expose students to “real” patients, facilitating their contact with the clinical context in university hospitals or partner institutions. This pedagogical approach will certainly enhance respect for sentient living beings, without compromising the established teaching-learning outcomes (Da Graça Pereira et al., 2017; Baptista, 2019).

This study presented several limitations. In fact, when optimizing the questionnaire, only MIMV students were included and most of MIM FMUP undergraduates stated that they did not attend classes involving animals (whether they were alive or cadavers), meaning that they were not able to answer questions number 3, 4 and 5. Additionally, the dissemination of the survey was heterogenous, namely, in the case of FMUP, the data collection occurred after an exam, which may have reduced the commitment of the students; in the case of MIM ICBAS students, the questionnaire was completed several days after having access to it, which resulted in a low response rate. Even when conducted in person, it was not always feasible for someone directly involved in the study to be present to provide potential explanations. The findings from this study cannot generalize to the entire Portuguese context as data was only collected in two medical and

two veterinary schools from the northern region of Portugal. A larger sample and wider coverage of other medical schools may, in the future, help to understand more deeply some of the aspects addressed here. Finally, the limited description of the procedures performed to the animals (open questions that require memory usage) did not always allow to determine their degree of severity.

The results included in this paper were developed in the context of a master's course and part of its findings were presented at a conference (Baptista et al., 2022).

Conclusion

In this study, nearly two thirds of students agree with sacrificing, breeding, maintaining and performing procedures on animals exclusively for educational purposes. Additionally, the majority of students holds a relational ethical profile (only less than one fifth agrees with the use of all species for educational purposes) and a utilitarian perspective towards animals. The latter implies that they consider the ethical acceptability of animal use in educational contexts based on the balance between benefits and costs, as long as there are measures in place to ensure the refinement of animal procedures. Most students are not aware of alternative methods that could replace animals in teaching, but most agree that a compromise must be achieved between learning objectives, teaching-learning methodologies and animal welfare/wellbeing. We believe that promoting academic integrity in the use of animals is the responsibility of educators, in order to contribute to the dissemination and implementation of teaching methodologies in life sciences higher education that are equally effective, but also more ethical, humane and compassionate.

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