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HOW WE TEACH | *Classroom and Laboratory Research Projects*

Effect of an educational game on university students' learning about action potentials

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Luchi KCG, Montrezor LH, Marcondes FK. Effect of an educational game on university students' learning about action potentials. *Adv Physiol Educ* 41: 222–230, 2017; doi:10.1152/advan.00146.2016.—The aim of this study was to evaluate the effect of an educational game that is used for teaching the mechanisms of the action potentials in cell membranes. The game was composed of pieces representing the intracellular and extracellular environments, ions, ion channels, and the Na^+-K^+-ATPase pump. During the game activity, the students arranged the pieces to demonstrate how the ions move through the membrane in a resting state and during an action potential, linking the ion movement with a graph of the action potential. To test the effect of the game activity on student understanding, first-year dental students were given the game to play at different times in a series of classes teaching resting membrane potential and action potentials. In all experiments, students who played the game performed better in assessments. According to 98% of the students, the game supported the learning process. The data confirm the students' perception, indicating that the educational game improved their understanding about action potentials.

teaching; educational games; active teaching methods; physiology; action potential

STUDENTS' LACK OF MOTIVATION and engagement is one of the major issues in education worldwide (2). Araújo et al. (4) observed a significant reduction in learning motivation levels between students of the second and eighth semesters of an undergraduate course in Psychology. Carmo and Carmo (12) also observed diminished student motivation during an undergraduate course in Accounting Sciences. Students with higher scores for interest and enjoyment, evaluated by the intrinsic motivation inventory, showed higher course grades (5, 15). Therefore, a higher level of intrinsic motivation has positive impact on learning performance.

In addition to the lack of motivation, uncertainty regarding knowledge of previously studied content was observed in undergraduate students (38), impairing their performance and problem-solving abilities (9). Inspired and motivated students seek knowledge in different ways and learn better (16).

To improve student motivation, engagement, and performance, the curriculum should change from a focus on the transmission of information by the teacher and memorization of content to active teaching-learning methodologies with active participation by the student, thus promoting effective learning. Active teaching-learning methods encourage the pursuit of information by the students themselves (41) and stimulate development of their autonomy, reasoning, and critical thinking (13, 35), by making the students the protagonists in their learning processes (22, 24). These methodologies favor the development of critical, thoughtful, professionals with better decision-making skills (28, 46).

There are different possibilities for encouraging student participation in the learning process, and it is up to the teacher to select pedagogical strategies according to the subject matter and teaching conditions (47). Educational games are an example of active teaching-learning methodologies that promote student engagement and contribute to their learning (19). The educational game is not necessarily a competitive activity and can be a simulation based on a real topic (3). This type of game allows students to solve problems by discussing the possible solutions with their peers, in play in an entertaining way (40, 43). Different games have been used in health courses to teach topics in obstetrics (31), psychiatry (6), immunology (18), pharmacology (8), and physiology (26, 27, 32, 37).

Although it has been reported that educational games had a positive impact on the teaching-learning process (27, 32, 37), Abdulmajed et al. (1) showed that the methodologies used were not totally reliable for measuring the level of learning. Flaws in the study design, game descriptions, sample sizes, variability of students and teachers, and in the assessment tool are examples of fails indicated by different authors (1–3). Therefore, it is important to improve the studies to investigate the effect of educational games on student learning.

In Physiology courses, students often have difficulty understanding the concepts of membrane potential and action potential. These topics require the teacher to link necessary concepts from different previously studied subjects, hindering the success of the teaching-learning process (7). Different strategies have been developed to teach these themes (10, 11, 29, 39, 45). Many studies have described models using animals or physiology laboratory materials to explain membrane and action potential. For example, Thurman (45) used frogs as a model, while Moran and collaborators (29) created a model to illustrate the generation of membrane potentials using materials found in the physiology laboratory, such as a dialysis membrane chamber, agar bridges, calomel electrodes, and others. The need to use animals or laboratories for these models makes it impossible to reproduce them in any teaching environment. On the other hand, some studies have presented models that are cheaper and easier to add in many educational settings. Cardozo (11) created a model using springs to explain the rela-

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tionship between ionic conductance and potential across the membrane. In 2016, the same author and colleagues (10) described an intuitive way to understand how membrane potential is established by the concentration gradient of K^+ ions, using simple principles familiar to high school graduates, as, for example, ions move randomly and charged ions are attracted by the opposite and are repelled by the same electric charge. Rodrigues-Falces (39) developed a model to explain the electrical behavior of the action potential in terms of elementary electrical sources, using graphical representations. All of these studies provided models to enable the students to reflect on the membrane and action potentials in different ways.

The coordinator of the present study had difficulty in understanding these concepts when she was an undergraduate student. Seeing the same problem in her students, she developed an educational game to make the concepts of membrane potential and action potentials more comprehensible. The educational game presented in this work has the advantage of being a low-cost teaching tool, easy to reproduce anywhere in the world, besides not requiring the use of a laboratory. In addition to allowing the students to have time to reflect on the lesson content, this educational game has the advantage of addressing the theme in an entertaining way, hence stimulating the students' interest in the topic. The purpose of this article is to describe this educational game concerning action potentials and evaluate its effect on students' learning.

MATERIAL AND METHODS

Educational Game

The material used in the educational game consisted of one sheet of white A3 size card (42 × 29.7 cm), with a red strip in the center (Fig. 1), and the following items: *1*) rectangular pieces of card indicating *media 1* and *2* (n = 1; 2 × 4 cm; Fig. 1); *2*) round pieces of card (2 cm in diameter; Fig. 1) in yellow, green, and pink, representing the ions Na^+ (n = 12), K^+ (n = 12), and Cl^- (n = 24), respectively; *3*) rectangular pieces of card indicating intracellular medium and extracellular medium (n = 1; 2 × 4 cm; Fig. 2); *4*) round pieces symbolizing negative proteins (n = 3; 4 cm in diameter); and *5*) rectangular pieces with figures of ion leak channels for K^+ (n = 1; 2 × 6 cm; Fig. 2*A*), Na^+ (n = 1; 2 × 3.5 cm; Fig. 2*B*), Na^+-K^+-ATPase pump (n = 1; 2 × 2.5 cm; Fig. 2*C*), closed voltage-gated channels of Na^+ and K^+ (n = 1; 2 × 4 cm; Fig. 2*D*), and voltage-gated channels of Na^+ (n = 1; 2 × 3.5 cm; Fig. 3*A*) and K^+ (n = 1; 2 × 5 cm; Fig. 3*B*), showing the open and closed states.

During the game activity, two teachers and four graduate students acted as monitors. The students were divided into groups (n = 4–5) and received a guide and the pieces of the game. The activity was divided into three levels.

Game level 1: Diffusion and equilibrium potentials. The students received the white card and the pieces (Fig. 1) indicating *environments 1* and *2* and the ions K^+ (n = 6), Na^+ (n = 6), and Cl^- (n = 12), to work on the concepts of diffusion and equilibrium potential. The students were asked to place the pieces in such a way as to show that, in *environments 1* and *2*, there were solutions with the same concentrations of NaCl and KCl, respectively, separated by a permeable membrane. They were told to move the pieces to show what the equilibrium state would be and were asked whether there would be movement of ions through the membrane in the equilibrium state. Next, the students were asked to move the pieces again, based on the same initial situation, while this time the membrane was only permeable to potassium. This sequence of procedures followed the explanation in the textbook previously provided for reading at home (42). The questions proposed for discussion were as follows: *1*) What is the direction of the forces generated by the concentration and electrical gradients?; *2*) What is the distribution of the ions when the two sides are in an equilibrium state?; and *3*) During this equilibrium state, will there be movement of ions through the membrane?

Game level 2: Resting membrane potential. In *level 2*, the concept of resting membrane potential was studied. The students were told to consider the two sides of the white card as extracellular and intracellular environments, separated by the cellular membrane. They received the pieces indicated in Fig. 2 to distribute within the extracellular environment (6 Cl^-, 5 Na^+, and 1 K^+) and the intracellular environment (3 Cl^-, 1 Na^+, 5 K^+, and 3 proteins). They also received pieces to position in the membrane: the K^+ (Fig. 2*A*) and Na^+ (Fig. 2*B*) leak channels, the Na^+-K^+-ATPase pump (Fig. 2*C*), and voltage-gated channels (in the closed state; Fig. 2*D*). The students were asked to move the ions, showing how the resting membrane potential was generated and maintained, explaining the role of the Na^+-K^+-ATPase pump in maintaining the resting membrane potential. The aim of the initial distribution described above was to illustrate the higher con-

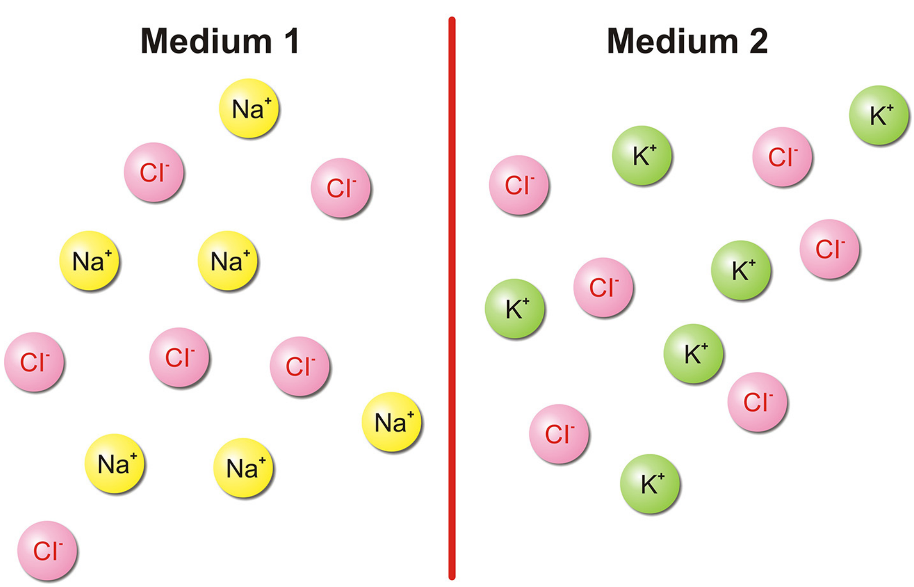

Fig. 1. Pieces of the educational game used in *level 1* of the activity on the concepts of diffusion and equilibrium potential.

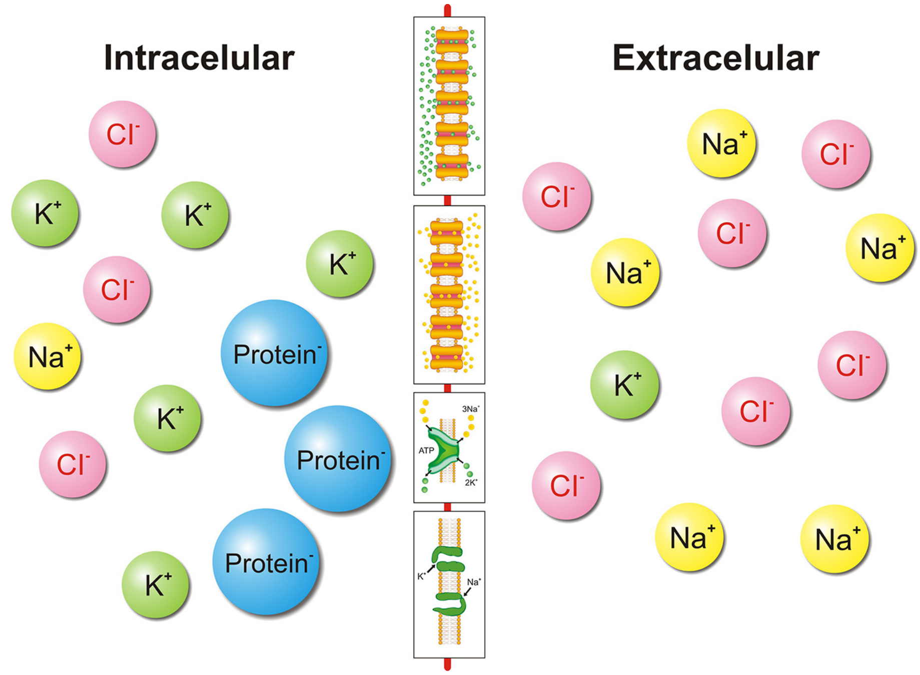

Fig. 2. Pieces of the educational game, representing ions and channels, used in *levels 2* and *3* of the activity, related to the concepts of resting membrane potential and action potentials, respectively. *A*: ion leak channels of K^+. *B*: ion leak channels of Na^+. *C*: Na^+-K^+-ATPase pump. *D*: closed voltage-gated channels of Na^+ and K^+.

centrations of Na^+ in the extracellular medium and of K^+ in the intracellular medium, and the presence of negatively charged proteins in the intracellular environment. The monitors explained that this was a simplified simulation of the intracellular and extracellular environments. *Levels 1* and *2* of the activity were used to support the content presented in *class 1* and studied in the textbook as homework.

Game level 3: The action potential. In this level, the groups received the voltage-gated channels pieces (Fig. 3) to be used with the pieces employed in *level 2*. By using the pieces indicated in Figs. 2 and 3, the students were asked to demonstrate the movement of the ions through the cellular membrane during an action potential. They were asked to show how the ion transport occurs during depolarization, repolarization, and hyperpolarization, and to indicate these changes in the resting membrane potential graphically. The groups discussed the events occurring during depolarization, repolarization, and hyperpolarization, considering whether the membrane potential was lowered or increased, and whether the cell become more positive or negative. Next, using an illustration of an action potential, the students were asked to indicate its phases and the state (open/closed) of the leak channels, voltage-gated channels, and the Na^+-K^+-ATPase pump (active or not) during the action potential.

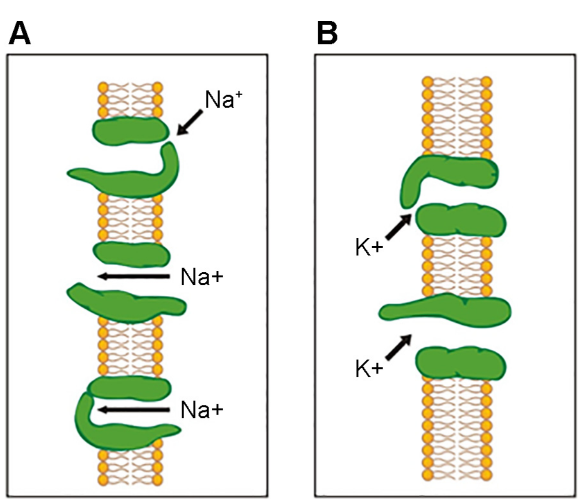

Fig. 3. Pieces of the educational game, representing voltage-gated ions, used in *level 3* of the activity, related to action potential concepts. *A*: voltage-gated channels of Na^+. *B*: voltage-gated channels of K^+.

The purpose of *level 3* was to teach the students the concept of action potentials by means of an entertaining group activity, based on the textbook reading they had done and on the concepts presented in the first lesson on resting membrane potential.

During the activity, the monitors checked if the pieces had been positioned correctly, without themselves making any corrections. If there were errors, the students were encouraged to identify them and then correct the positioning of the pieces, guided by the monitors' questions and group discussion. This activity lasted around 90 min.

Procedures

First-year students of the undergraduate course in Dentistry at the Piracicaba Dental School of the University of Campinas (FOP-UNICAMP), who took the subject of Biosciences I in 2015 (*experiment 1*; $n = 67$) and 2016 (*experiment 2*; $n = 81$) were invited to participate in this study. When enrolling in the undergraduate program, the students were randomly divided into two groups by the academic system: *A* and *B*. Based on this division, at different times they attended practical lessons in laboratories that could accommodate up to 40 people. For the present study, the two groups were randomly assigned to either the control group or the game group.

The aim of this study was to determine whether students who used the game performed better than students who did not use the game. In the first experiment, students in the game group completed an assessment immediately after undertaking the game activity. To avoid the influence of the immediate effect of the game, in the second experiment, the assessment was performed 2 days after the group had completed the game. The sequences of procedures used in *experiments 1* and *2* are presented in Fig. 4.

Experiment 1. CLASS 1. In *class 1*, all of the students attended a 75-min theoretical class on resting membrane potential. To spark the students' interest, the teacher asked them to form pairs to discuss the following questions: *1*) How do local anesthetics act? and *2*) How are electrical stimuli transmitted in the body? After 10 min of discussion, the students were invited to present their answers, and the teacher

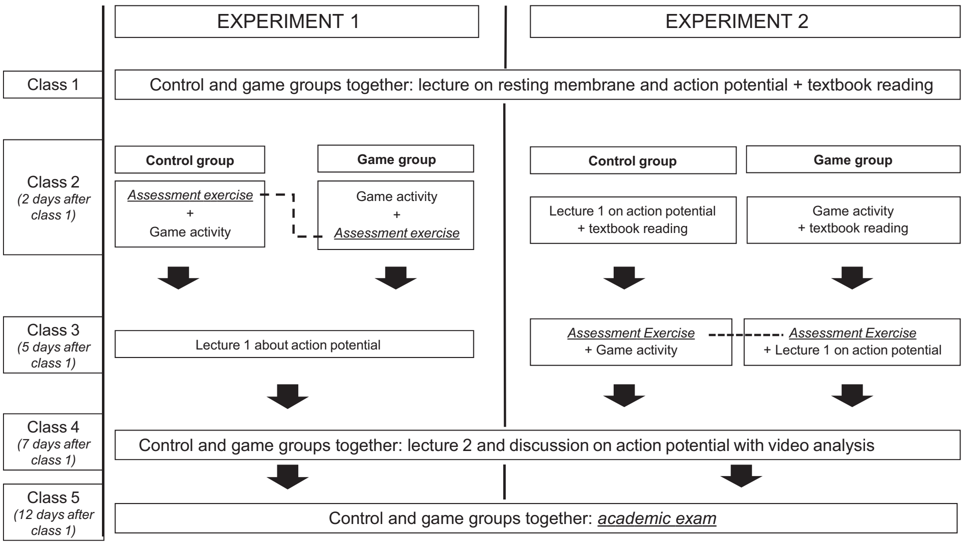

Fig. 4. Sequence of procedures used in the experimental designs of *experiments 1* and *2*.

informed them that these topics would be studied in the four following classes.

Next, the teacher used PowerPoint slides to present the topics outlined in Table 1 (*lecture 1* on resting membrane potential). In this lecture, the teacher explained that the distribution of electrical charges through the membrane could be modified to promote cell depolarization, repolarization, and hyperpolarization. However, the participation of voltage-gated channels was not explained. The objective was to show the students that the transmission of a nerve impulse occurs through depolarization of the cell and rapid repolarization, so that the cell can promptly respond to the new stimulus. At the end of this class, the students were instructed to study the topics of membrane potential and action potential in the textbook (42), in preparation for an assessment that would take place during the following class, 2 days later.

CLASS 2. At the beginning of *class 2*, the control group was informed that the assessment would be an exercise that would not count toward the subject grade, with the goal being to evaluate the students' learning progress so that the teacher could assess whether any of the items had to be reviewed before proceeding to the next topics. After this assessment exercise about resting membrane potential and action potential, the students performed the activity with the educational game. The game group took the same assessment immediately after performing the game activity. The game was used as a reinforcing activity for membrane resting potential and as an active teaching-learning method for action potential.

CLASS 3. In *class 3*, both groups received a lecture about action potentials, with a discussion about the game activity from the previous class and the questions of the assessment exercise. The teacher presented slides explaining the topics described in Table 1 (*lecture 1* on action potential).

CLASS 4. In *class 4*, there was a second lecture about action potentials, with the presentation and analysis of a video obtained from the Portuguese Society of Neurosciences (www.spn.org.pt - Free material for Brain Education / Films / Action Potential and Epilepsy), concerning the transmission of nerve stimuli in epilepsy. This video showed the behavior of the ion channels during the resting membrane potential and during one action potential, as well as a graph of the action potential. The students were instructed to watch this video in pairs and identify the ion channels in the plasma membrane and the phases of the action potential. Using PowerPoint slides, the teacher showed the relationship between the states of these channels and the graph of the action potential, following the topics described in Table 1 (*lecture 2* on action potential).

CLASS 5. In *class 5*, the students took the academic exam with questions about resting membrane potential and action potentials.

Experiment 2. CLASS 1. The same procedures described for *experiment 1* were followed, and the same topics were addressed.

CLASS 2. The control group attended a lecture on the action potential, addressing the topics indicated in Table 1 (*lecture 1* on action potential). The game group performed the educational game activity without previous lecture on action potentials.

At the end of this class, all of the students were instructed to study the action potentials topics in the textbook (42), to prepare for an assessment that would take place during the following class, 3 days later.

CLASS 3. At the beginning of *class 3*, both groups took an assessment exercise about action potentials. After this assessment, the control group performed the game activity, and the game group attended the lecture on action potentials, addressing the same topics

Table 1. *Topics discussed in the lecture sessions in experiments 1 and 2*

Topics Discussed
Lecture on resting membrane potential
• Composition of the intracellular and extracellular environments.
• Diffusion of substances through permeable and semipermeable membranes.
• Permeability of the plasma membrane and its relationship to ion leak channels.
• Differences in electrical charge generated by a selectively permeable membrane.
• Equilibrium potential of an ion and the Nernst equation.
• Difference in electrical charges through the plasma membrane.
• Sodium (Na^+) and potassium (K^+) leak channels present in the plasma membrane.
• Goodman-Hodgkin-Katz (GHK) equation.
• Function of the Na^+-K^+-ATPase pump.
Lecture 1 on action potential
• Dynamics of voltage-gated ion channels.
• State of leak channels during phases of the action potential.
• Difference between graded potentials and action potentials.
• Graph of the action potential over time.
• State of ion channels during different phases of the action potential.
Lecture 2 on action potential
• Relative and absolute refractory periods.
• Influence of changes in the ion concentrations in the extracellular environment on triggering action potentials.
• Action potential mechanisms and drug development.

considered in *class 2* for control group (Table 1, *lecture 1* on action potentials).

CLASSES 4 AND 5. The same procedures described for *experiment 1* were followed.

Learning Assessment

The students' learning progress was assessed in an exercise performed in *class 2* (*experiment 1*) or *class 3* (*experiment 2*), as well as by the answers to the questions in the academic exam performed in *class 5*. The learning assessment questions used in *experiments 1* and *2* were not exactly the same, but they required the same knowledge about the themes studied. Thus, even if students contacted colleagues and knew the questions used in the previous year, the answers to the questions could not be the result of memorization.

In *experiment 1*, the assessment consisted of four questions. *Question 1* was a multiple choice on resting membrane potential. In *question 2*, the students had to identify the regions of depolarization, repolarization, and hyperpolarization on a graph, indicating the resultant flux of charges through the membrane (entry or exit of positive or negative charges into and out of the cell) and determining whether there was an increase or decrease in the resting membrane potential. In *question 3*, the students were asked to explain the role of the Na^+-K^+-ATPase pump in the membrane potential. In *question 4*, the students were required to analyze an action potential graph and indicate the state (open or closed) of the leak channels and the voltage-gated Na^+ and K^+ channels, as well as the activity of the Na^+-K^+-ATPase pump (active or not). *Questions 1–3* were used to analyze learning about resting membrane potential, and *question 4* was used to evaluate learning about action potentials.

In *experiment 2*, the assessment was composed of two multiple-choice questions about action potentials. In *question 1*, the students had to analyze a graph of action potentials and identify the regions of depolarization, repolarization, and hyperpolarization, indicating the resultant flux of charges through the membrane (entry or exit of positive or negative charge into and out of the cell). In *question 2*, the students were asked to indicate the state (open or closed) of the leak channels and the Na^+ and K^+ voltage-gated channels, and the activity of the Na^+-K^+-ATPase pump (active or not). In both experiments, the students were instructed to leave the questions they were unsure of unanswered. Thus it was possible to identify the topics the students knew they did not understand (unanswered questions) and those that they had understood incorrectly (incorrect questions). For this study, the exercises were scored using a value from 0 to 10; however, the score obtained was not considered for the subject grade. The hypothesis was that the students who did the exercise after the game would perform better in the assessment, compared with the control group.

In *class 5*, all of the students took the academic exams of the Biosciences I subject. Since this was an integrated subject, the exam included questions on anatomy, histology, biochemistry, and physiology. The academic exam consisted of 28 questions in *experiment 1* and 25 questions in *experiment 2*, with 3 questions being on histology, 12 (*experiment 1*) or 9 (*experiment 2*) on biochemistry, and 13 on physiology in both experiments. The physiology questions were divided as follows: one about homeostasis, eight about blood, and four about resting and action potentials. To evaluate the effect of the game, the four questions about resting and action potentials were considered in this study. These questions covered similar topics of the assessment exercise, but were not the same questions. In the academic exams, each question was worth 0.36 points (*experiment 1*) or 0.40 points (*experiment 2*). To compare the performance of the groups in this study, in *experiment 1*, the grades obtained in these questions were normalized as if they were worth 10 points. In *experiment 2*, only the questions about action potential were considered and were normalized in the same way.

The aim of the scores obtained in the academic exam questions was to evaluate whether the sequence of activities affected the medium-term learning ability of the students. By the time of the academic exam, both control and game groups had performed the educational game activity, so our hypothesis was that, if the game was effective, irrespective of whether it was used before or after the lectures on action potential, there would be no significant difference between the performances of the two groups. In addition, this evaluation aimed to determine whether there was a difference between the groups. If no difference was observed in the academic exam, this would suggest that the difference observed in the assessment exercise was not due to differences between the groups, but was probably related to the procedures of the experiments. The same teacher, who was blinded to the groups, marked all of the answers in the assessment exercises and academic exams.

Students' Perception about the Educational Game

After the academic exam, the students were asked to answer a question on whether the game helped them in their learning, and to explain the reason for the answer provided. This evaluation aimed to assess the students' perception of the game.

Ethical Approval

This study was approved by the institutional ethics committee (protocol no. 22/2015). Only students who participated in all of the lessons and who agreed to participate and signed the Term of Free and Informed Consent were included in the study.

At the end of the semester, after receiving their final scores, the students were informed of the study and were invited to participate, authorizing the use of their responses in the assessment exercise, the theoretical test, and the question concerning their perception of the game. This was performed to minimize vulnerability of the participants and ensure their autonomy (44), since they were students of the researcher responsible for this study.

Statistical Analysis

For statistical data analysis, the unpaired Student's *t*-test (5% significance level) was used to compare the control and game groups in terms of numbers of incorrect answers, unanswered questions in the assessment exercises, and the grades obtained in the exercises and in the academic exams.

RESULTS

Sixty-one and sixty-six students, out of classes of 67 and 81 students, signed the informed consent and participated in *experiments 1* and *2*, respectively.

In *experiment 1*, the aim was to evaluate the effect of the game on learning about resting membrane potential by comparing the group that attended a lecture and studied the topic at home (control group) and the group that not only attended the lecture and studied at home, but also performed the activity with the educational game (game group). Considering the learning about action potentials, the group that studied the subject by reading the textbook (control group) was compared with the group that, in addition to reading, performed the activity with the educational game (game group). Therefore, the comparison was made between the effects of reading and game activity on students' learning about action potential. In the assessment of *experiment 1*, the game group students had higher scores and fewer mistakes, compared with the control group in the questions about resting membrane potential (Table 2; $P < 0.05$), without any difference in the number of unanswered questions (Table 2; $P > 0.05$). For the questions about action potentials, the game group had higher scores and fewer errors and unanswered questions, compared with the control

Table 2. *Students' performance in the assessment exercise on resting membrane potential and action potentials in experiment 1*

Group	*n*	Grade	Mistakes	Unanswered Questions
Resting potential				
Control (reading)	31	6.41 ± 0.35	2.77 ± 0.31	1.77 ± 0.41
Game	30	7.86 ± 0.28*	1.57 ± 0.26*	0.86 ± 0.20
Action potential				
Control (reading)	31	4.75 ± 0.32	5.00 ± 0.45	2.84 ± 0.62
Game	30	8.09 ± 0.45*	1.93 ± 0.38*	0.93 ± 0.47*

Values are means ± SE; *n*, no. of observations/group. Grade shows the sum of values obtained in questions answered correctly (total or partially). Mistakes shows the no. of questions answered incorrectly, indicating errors in student comprehension. Unanswered Questions shows the no. of questions that were not answered, indicating the information and concepts that the students realized they did not understand. *Significantly different from control group (Student's *t*-test, $P < 0.05$).

group (Table 2; $P < 0.05$). There were no differences between groups in terms of the grades obtained in the academic exam for questions on resting membrane potential (game group: 8.02 ± 0.30; control group: 8.15 ± 0.31; $P > 0.05$) and action potentials (game group: 7.25 ± 0.40; control group: 7.34 ± 0.36; $P > 0.05$).

In *experiment 2*, learning about action potential mechanisms was evaluated by comparing the performances of the group that attended a lecture and studied the topic at home (control group) and the group that studied the topic at home and performed the game activity (game group). Therefore, the comparison was made between the effects of the lecture and the game activity on students' learning about action potentials. The group that performed the assessment after the game activity had higher scores and fewer mistakes, compared with the control group that attended a lecture (Table 3; $P < 0.05$). There were no differences in the number of unanswered questions (Table 3; $P > 0.05$) or in the group scores obtained in the academic exam (game group: 7.25 ± 0.40; control group: 7.34 ± 0.36; $P > 0.05$).

According to nearly all (126 out of 127) of the students who answered the question to assess the students' perception of the game, the game helped them learn. According to these students, the game clarified the content and helped them understand it ($n = 31$); helped them visualize abstract content ($n = 29$); was a fun and dynamic learning method ($n = 19$); and helped link the topics ($n = 13$). Only one student judged that the game did not help him/her learn, because he/she preferred moving slides. Some students did not explain their answers.

DISCUSSION

The results of this study indicated that the use of the educational game enhanced the university students' learning about resting membrane potential and action potential. The students also observed that the game had a positive effect.

Although educational games have been used to teach different subjects, there is no consensus in the literature concerning their effectiveness in learning. Some studies showed that educational games improved learning (8, 14, 37), while others did not (17, 27). According to Barclay et al. (8), randomized studies are needed that compare knowledge acquired by using an educational game and that obtained by means of theoretical lessons.

The present study was developed in the context of these uncertainties and requirements. In *experiment 1*, comparison was made of the performance of students in an assessment exercise about resting membrane potential, after use of the combination of the educational game, lecture, and textbook reading, and after the lecture and textbook reading only. Considering action potentials, comparison was made between performance after textbook reading and after textbook reading plus the educational game activity. In *experiment 2*, the performance of the students after use of the educational game was compared with the performance observed after a lecture.

It is important to highlight that, in the case of the assessment exercise, the unanswered and incorrect answered questions were evaluated. The results indicated that, in the game group, there were fewer concepts and details understood incorrectly (incorrect answers), compared with the control group, without difference in concepts that the students realized that they had not understood (unanswered questions).

These findings suggested that the game, used to reinforce the theoretical lesson, may have helped to clarify the concept of resting membrane potential, making it easier for the students to understand the subject. The results of the students' opinion of the game supported this interpretation, since 99% of the students reported that the game helped them to learn because it made the content easier to grasp and the abstract concepts more perceivable as reality.

For the concept of action potentials, the comparison proposed in *experiment 1* was between the textbook reading and the textbook reading plus the educational game activity. The basic content on membrane potential, explained in the theoretical lesson, should have enabled the student to understand the subjects of the assigned reading regarding action potentials. However, the results obtained showed that this did not in fact happen. The better performance of the game group in the questions on action potentials confirmed that the students had difficulty with this topic, since the control group answered fewer questions correctly.

The purpose of this comparison was to evaluate whether the game would be effective as a tool for promoting the learning of action potentials. During the activity with the educational game, some students spontaneously said that the game was helping them to understand the material that they had read in the textbook. They said that some topics seemed very abstract and that by manipulating the pieces of the game, they became more understandable. Similarly, the activity with the game helped the students to understand the dynamics of the ion channels demonstrated in the video used in *class 4*. During this

Table 3. *Students' performance in the assessment exercise on action potentials in experiment 2*

Group	*n*	Grade	Mistakes	Unanswered Questions
Control (lecture)	34	7.19 ± 0.29	7.62 ± 0.82	0.53 ± 0.18
Game	32	8.16 ± 0.24*	4.81 ± 0.67*	0.59 ± 0.20

Values are means ± SE; *n*, no. of observations/group. Grade shows the sum of values obtained in questions answered correctly (total or partially). Mistakes shows the no. of questions answered incorrectly, indicating errors in student comprehension. Unanswered Questions shows the no. of questions that were not answered, indicating the information and concepts that the students realized they did not understand. *Significantly different from control group (Student's *t*-test, $P < 0.05$).

lesson, the students spontaneously said that, when analyzing the video, they remembered how they had moved the ions during the educational game activity.

A limitation of this analysis was the absence of a comparison between the scores obtained by a group that used the educational game, and another that attended a lecture in which all the information was transmitted by the teacher. Another limitation in *experiment 1* was that the effect of the educational game was evaluated immediately after the game activity.

Therefore, *experiment 2* was designed with the aim of comparing the midterm effects of a lecture about action potentials and the activity with the educational game, in terms of the students' learning ability. Higher grades and fewer mistakes were recorded for the game group, supporting the conclusions of *experiment 1* and demonstrating the better performance of the students after the activity with the educational game, compared with a lecture. Moreover, the assessment exercise was performed 2 days after the activity with the game, so the effectiveness of the latter in improving the understanding of membrane potentials was not restricted to immediate effects.

This interpretation was reinforced by the absence of any difference between the performances of control and game groups in the academic exams in *experiments 1* and *2*. The academic exams were held 12 days after the two groups had undertaken the activity with the educational game. In the future, we intend to conduct complementary studies to analyze the effect of the educational game on long-term learning, with assessment of what the students can still remember about action potentials and the generation and transmission of electrical stimuli when they reach the second to fifth years of the undergraduate course.

The positive effect of this game may be related to factors involved in adult learning, whereby students feel motivated to learn when they are able to integrate the content studied with their professional activities, when they can apply the new content quickly, and when they feel responsible for their own learning (25). In *class 1*, the relationship was shown between the topics that would be studied and the clinical practice of dentistry (involving local anesthetic and the transmission of nervous stimuli, including painful stimuli). During the game activity, the monitors asked questions so that the students could connect the basic concepts regarding the intracellular and extracellular environments, including the types of ion channels present in the cellular membrane, to the transport of ions through the membrane and generation of electrical stimuli. Although they were not measured, satisfactory reactions and even joy could be identified in some students when they realized that they had understood the subject by answering the questions correctly, or when they had offered an explanation of an aspect of the subject to another student.

In addition, during the series of lessons, repetition of concepts and content was made using different approaches: theoretical explanations with PowerPoint slides, reading, video analysis and discussion, and group discussions during the activity with the educational game. This allowed the students to have time to assimilate the information, connect it to previously studied content, and identify their doubts (25). Furthermore, the use of different strategies allowed the teacher to interact with the students employing varied sensory learning styles (20). The VARK questionnaire (where V is visual, A is aural, R is reading-writing, and K is kinesthetic) is a tool that can be used by teachers to determine the learning style preferences of each student. The VARK questionnaire is available in the link: http://www.llcc.edu/student-services/cas/helpful-handouts/using-the-online-vark-questionnaire/. People have different learning preferences, such as reading and writing, watching films or demonstrations, and engaging in activities involving movement and social interaction (36). Therefore, the use of an educational game associated with theoretical lessons and reading is an important strategy that can enhance the students' learning processes.

Another factor that may also have contributed to the positive effect of the educational game was the formative assessment, which provided the student with constant feedback (23). Throughout the activity with the educational game, the students needed to review the movement of the pieces representing the ions through the ion channels. The exchange of ideas during the group discussion to find the correct answer allowed the students to receive feedback from their colleagues or monitors. And these process could promote higher engagement of students and improve learning (21, 30, 33, 34).

The development of the game used here stemmed from the students' difficulty in understanding the subject, as identified by the teacher coordinating the study, who had experienced similar difficulty when she was an undergraduate student. During a conference, in a discussion about the preliminary data of the present study, another teacher reported to have also developed a game on the same topic, involving the movement of pieces simulating ions. It was evident from the exchange of information that the games seemed to help the students to learn and improved their levels of interest and engagement. In addition, it was noted that development of teaching strategies by teachers involves reviewing their own experiences as students, putting themselves in the place of their students. The sharing of experiences with other teachers is of great importance in establishing innovative teaching practices, because it allows teachers to perceive that their colleagues have the same difficulties, concerns, and questions, and that students of different courses and institutions present similar demands and difficulties.

Educational games employing structures and materials similar to those used here could be developed to teach other topics. The game was based on the use of pieces produced from cheap and simple materials to make the students think about the information they had acquired in the lecture and from reading a book. The pieces were designed to make the students use this information (concerning the cell membrane structure, ion channels, and ion concentrations inside and outside the cell) to understand how the movement of ions occurs and how the electrical charge differences through the cell membrane are generated and sustained. The different levels of the game allowed the students to understand the themes step by step. This approach could be used to teach other complex topics in physiology as well as in different areas.

It is important to stress that the present study evaluated learning not only considering the perception of the students, but also analyzing the scores obtained in the postgame assessment, as well as the numbers of errors and unanswered questions. Therefore, this study represents an important contribution concerning the influence of educational games on student learning, demonstrating their effectiveness.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the author(s).

AUTHOR CONTRIBUTIONS

K.C.G.L. and F.K.M. performed experiments; K.C.G.L., L.H.M., and F.K.M. analyzed data; K.C.G.L., L.H.M., and F.K.M. interpreted results of experiments; K.C.G.L. and F.K.M. prepared figures; K.C.G.L., L.H.M., and F.K.M. drafted manuscript; K.C.G.L., L.H.M., and F.K.M. edited and revised manuscript; K.C.G.L., L.H.M., and F.K.M. approved final version of manuscript.

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