Learning to understand complex biological systems by computer modeling and molecular mechanistic reasoning

Abstract

Students experience difficulties when they try to understand complex biological systems. When asked to explain a phenomenon, most students are not able to connect the different organizational levels and they tend to reduce the complexity of the system, focusing only on one structure instead of seeing the system as a whole. In this paper it is investigated how students can be guided to explain biological systems as a mechanism during a computer modelling task and how this combination of guiding their reasoning and computer modelling leads to a better understanding of the system. A new SimSketch application was designed to model the electrical properties of a nerve cell. Eight preuniversity students tested out this new software while they were guided to reason mechanically. Detailed analysis of students' conversations, modelling actions and filled-in worksheets revealed that it was possible to promote a mechanistic reasoning style during the modeling task and that this helped the students to gain deeper understanding of the target phenomenon. Hence, this exploratory research suggests that combining computer modelling with promotion of mechanistic reasoning is a powerful educational method to understand complex biological systems.

Introduction

In preuniversity biology education students are expected to understand some very complex systems and processes, such as photosynthesis, protein synthesis, and neural signaling. Key characteristics of complex systems include multilevel organization, interconnections, heterogeneous components, and invisible dynamic processes. These features make it challenging for novices to work with complex systems, because they are not able to connect the different organizational levels and see the system as a whole. Instead, when explaining a complex system, for example the resting potential of a neuron, novices often reduce the complexity, focus merely on one structure and wrongly assign the resting potential only to the sodium-potassium pump. Experts, on the other hand, are aware of the structures, behaviors and functions of the whole system (Hmelo-Silver, Marathe, & Liu, 2007).

The quality of an explanation of a biological phenomenon is linked to the question one can ask when approaching such a complex system. One can ask: 'what?'. This question is easily answered by summing up all the stuff that is in the system. 'Why?' will lead to an explanation focused on the function of a system and its components. Although this question is very useful, it might lead to so-called teleological or anthropomorphic explanations. Such explanations attribute goal-directed or intelligent behavior to lifeless objects, like ions and proteins. 'How?' is the most sophisticated question, because one has to unravel the mechanism underlying the phenomenon in order to answer it (Hmelo-Silver, Marathe, & Liu, 2007).

In preuniversity biology education, there is a tendency to focus on the structures and the functions of a system. The unfamiliarity with 'how-questions' and a tendency to ask 'why-questions' prevents students to dig deeper for real causal relationships, and results in teleological and anthropomorphic explanations of physical mechanisms (Kampourakis & Zogza, 2008, Abrams & Southerland, 2001). In the biological sciences important discoveries were often made based on teleological assumptions.

For example, biologist have searched for benefits of evolutionary adaptations, assuming that the species adapted towards a goal (Zohar & Ginossar, 1998). Nevertheless, this approach refrains students from zooming in, searching for a meaningful explanation and thus teachers and educational designers must be aware of the misconceptions that result from such an approach.

Another, but related problem is that students are not aware of the fact that interactions between proteins and other molecules and atoms explain higher-order phenomena and cell activities. For example, the motion of little ions over cellular membranes through ion channels and ion pumps are responsible for the electric potential that cells use for signaling and transport of other particles. At first sight ions Students often do not recognize the multiple organizational levels between the molecules and cells, and therefore do not understand the relationship of molecules and cells. Furthermore, students are not aware how proteins and their interactions are organized in time and space, so they are not able to develop a holistic view of biological systems (van Mil, Boerwinkel, & Waarlo, 2013).

So, understanding complex biological systems is difficult for students due to a lack of knowledge of molecular interactions that are the basis of these systems. Also, students need to learn to apply the proper reasoning. In order to connect the dots, students need a combination of insights in the nature of molecular interactions and scientific reasoning skills. Van Mil et al. (2013) described the skill to explain (complex) biological phenomena by referring to molecular interactions in a mechanistic way as 'molecular mechanistic reasoning'.

Molecular mechanistic reasoning is a domain-specific version of mechanistic reasoning, described in literature as leading to meaningful explanations of natural phenomena. It is "nonteleological (…) causal (…) built from experience (…) and it describes underlying or relevant structures" (Russ, Scherr, Hammer, & Mikeska, 2008, pp. 503-504).

Molecular mechanistic reasoning is different from general mechanistic reasoning because it involves domain specific knowledge about the behavior of (biological) molecules, such as the fact that proteins collide, bind, and change shape in order to fulfill their function. It is characterized as "hypothesizing, constructing and interpreting mechanistic explanations for (sub) cellular phenomena, while taking into account the physical and chemical principles that drive changes at the bottom level of molecular interactions" (Van Mil, Postma, Boerwinkel, & Waarlo, 2016). In summary, it is a scientific type of reasoning that chains protein interactions into a mechanistic model, thereby giving a mechanistic explanation for biological phenomena. Molecular mechanistic reasoning is less intuitive than mechanistic reasoning because molecular interactions are harder to understand in comparison with visible systems we are familiar with. For example, it is easier to explain mechanically how a car engine works than it is to explain mechanically how a nerve cell maintains its resting potential.

Van Mill et al. designed a lesson series in which secondary school students are guided to reason mechanically about several biological systems. In this study, students learned strategies based on the strategies scientists use for explaining biological phenomena. The first strategy is a top-down approach, in which students learn to ask how-questions, subdivide the phenomenon and hypothesize mechanistic schemas. In the second phase, students learn to 'explore the bottom level' by chaining molecular interactions into a mechanistic schema. In the third phase, students 'zoom out', combining the acquired insights from the bottom level into a model that explains the studied phenomenon.

Computer models could help to make these last two steps more efficient, because by computer modeling students are able to model a phenomenon without keeping all the variables and parameters 'in their heads', thereby offloading memory (Löhner, van Joolingen, & Savelsbergh, 2003). Also, students can easily change structures, parameters, interactions etc. and thereby exploring the system by inquiry (Rutten, van Joolingen, & van der Veen, 2012). Moreover, there is a resemblance between the use of (computer) models and mechanistic reasoning because they are both methods to represent a phenomenon in a way that goes beyond merely summarizing empirical discoveries. Both are scientific methods that focus on the mechanics behind a phenomenon and make us able to transfer this knowledge to other related contexts and to make predictions. Successfully creating a model always goes together with reasoning about the underlying mechanism. Therefore, it is natural to actively stimulate mechanistic reasoning during a modeling task.

The effectivity of modeling in secondary science education has been evaluated in several studies (Louca & Zacharia, 2012). One of these studies tested an inquiry-oriented physics curriculum based on modeling for middle school students. The authors showed that both inquiry skills and knowledge about physics improved significantly during the experiment (Schwartz & White, 2005). A design based-study implemented modeling of systems biology in biology education. The authors conclude that visualizing the cellular processes and the relation between different organizational levels of biology helps students to acquire a coherent understanding of the processes of cell biology (Verhoeff, Waarlo, & Boersma, 2008). This research is a strong indication that modeling activities may have great potential for learning complex biological systems.

The biological topic for this study is the membrane potential of a nerve cell: the resting potential and the action potential. This subject was chosen by the first author because he, as a biology teacher, observed that most student experience this topic as very challenging. This leads to common misconceptions regarding neural signaling. An example, derived from website of the Dutch Open University in which common misconceptions are documented, is that students often think that neurotransmitters can only *stimulate* neurons, neglecting that they can also be inhibitory (Kennisbank misconcepten in de biologie). A neural membrane also corresponds with the previously mentioned characteristics of complex biological systems as outlined by Hmelo-Silver, Marathe & Liu (2007): there are multiple levels of organization (tissue system, cellular and molecular), interconnections (the membrane potential is connected to permeability of the membrane for specific ions and vice versa), heterogeneous components (multiple ions and membrane proteins, membrane itself etc.) and invisible dynamic processes (like the change in membrane potential). Also, it is difficult to hypothesize a meaningful explanation of these phenomena without knowledge of how proteins perform their tasks and how ions influence a membrane potential, making this topic suitable for studying (molecular) mechanistic reasoning.

The aim of this study is to explore how students can gain deep understanding of complex biological systems by using computer modeling and mechanistic reasoning together. This leads to the following research question:

What are the properties of an effective learning trajectory for understanding complex biological systems in which computer modeling and stimulation of mechanistic reasoning are combined?

This question can be subdivided into three questions:

1. How can mechanistic reasoning be stimulated during a computer modeling task?
2. Does application of mechanistic reasoning lead to better performance on modeling tasks?
3. Does working on a modelling task lead to better reasoning?

Promoting molecular mechanistic reasoning in secondary biology education was only tested in an artificial research setting and with a series of lessons (Van Mil, Postma, Boerwinkel, & Waarlo, 2016). It has not yet been tested in schools, with less time and combined with computer modeling. There are a lot of unknown variables that have not been tested before. Moreover, we need new learning goals that focus on explaining a phenomenon mechanically. Thus, this exploratory research asks for a design-based research approach. This gives space to change the learning trajectory when, for example, students do not demonstrate mechanistic reasoning at all, have too little time for the tasks etc.

Design of the modeling assignment

As worked out in the introduction, an efficient method for understanding complex biological systems might be the combination of computer modeling and domain-specific mechanistic reasoning techniques. This hypothesis will be tested by developing and evaluating a computer modeling task during which students are encouraged to use mechanistic reasoning. The target phenomenon of this lesson is the physiology of a neural membrane. The learning goals are:

1. Learning to propose an explanation of how a neuron manages its membrane potential by actively or passively transporting ions over its membrane by membrane proteins in which higher-order mechanistic reasoning is used.
2. Learning to propose an explanation of how an action potential is generated and moves along an axon in which higher-order mechanistic reasoning is used.

The reasoning style that we try to promote in this study is inspired by molecular mechanistic reasoning but it is not exactly the same. Molecular mechanistic reasoning is more specific: it focusses on how changes on the molecular level, like conformation changes of proteins, explain higher-order phenomena. In our modelling software, which is further explained in the next paragraph, the molecular properties of proteins are kept ‘inside the black box’. Instead, we focus on the movement of the ions in order to explain the target phenomenon.

SimSketch

A good example of modeling software that allows students to experiment is SimSketch. SimSketch is a digital modeling tool in which specific tasks or behavior can be assigned to hand-drawn objects. “... SimSketch bridges the gap between informal, sketch-based representations and formal, executable models” (van Joolingen, Bollen, Leenaars, & Gijlers, 2012, p. 690). Modeling task being supported by free-hand drawing is a unique feature of SimSketch. Free-hand drawing supports modeling in two different ways: drawing a sketch to prepare the model and drawing the model itself. SimSketch combines the two and makes it possible to draw a functional model (van Joolingen, Bollen, Leenaars, & Kenbeek, 2010). Moreover, students do not have to learn specific modeling or programming skills to use SimSketch. Instead, they can directly and intuitively start with making models. The use of SimSketch is already tested in different educational settings and with different groups of learners. Research has shown that drawing supports the modeling process by activating prior knowledge by undergraduate students (Leenaars, van Joolingen, & Bollen, 2013). Another study confirmed that SimSketch contributes to learning of primary school children (Bollen & van Joolingen, 2013), which emphasizes the user-friendly nature of SimSketch. SimSketch has been used for biological topics in secondary education, such as predator-prey systems and swarming behavior. For the current research, SimSketch was adapted so that students can model a neuronal membrane by drawing elements like ions, ion-channels and ion-pumps.

Designing an effective educational computer modeling task

From research on drawing as a support for modeling activities it was learned that students experience modeling as a challenging task and thus need support. Difficulties students encounter are: they see the model as an artefact that has to work instead of dealing with it as a learning tool, they forgo prior knowledge and do not implement their knowledge into the model, and after using a modeling tool students neglect variables that were not represented in the simulation. The main suggestion for supporting students from this study is to let students draw an informal model before letting them work with the computer model. This prevents students from focusing on the computer model, thereby neglecting their prior knowledge and variables that are not represented in the simulation (Leenaars, van Joolingen, & Bollen, 2013). Another study, in which students' modeling performances and working methods were compared, gained insight in which methods students apply lead to sophisticated models, and how to scaffold the modeling process in order to let most of the students create a good model. From this study it was learned that successful students used a top-down approach, thereby seeing the model as a whole instead of a collection of separate variables, use inductive reasoning (students hypothesize about the interaction of model elements and behaviors) and use their prior knowledge. Implications for scaffolding modeling tasks include: activating prior knowledge, preferably in a collaborative setting, articulation of modeling subgoals, revising one model parameter at a time and stimulating a top-down approach (Sins, Savelsbergh, & van Joolingen).

These suggestions are implicated in the lesson design in the following way:

- Students draw an informal model of the resting membrane potential in dyads before they create a model in SimSketch.
- The modeling task is divided into different stages (modeling sub goals) with increasing complexity: first students learn to use the software, learn about the behavior of ions an how movement of ions influence membrane potential, subsequently they learn the influence of membrane proteins on ion flow and membrane potential, in the end students have to model an action potential with only little support.
- During the different students write down hypotheses about what will happen with the membrane potential. This might stimulate students to apply a top-down approach because the membrane potential is the result of movement of ions through membrane proteins, and stimulate students to use inductive reasoning (which is related to mechanistic reasoning).
- Students are stimulated (on the worksheets and by the teacher/researcher) to revise one parameter at a time.
- After the modeling activity, students are asked to think about elements of the system that are not simulated in the computer model.

Supporting inquiry learning

The lesson design gives students the possibility to experiment and construct their own knowledge by inquiry. Inquiry learning is widely accepted as an effective educational approach which gives students an authentic scientific experience and provides deep understanding, but it is also clear that students do not automatically perform inquiry in a scientific sound way (Kuhn, Black, Keselman, & Kaplan, 2000). Research on inquiry by computer modeling has shown that the scientific reasoning steps hypothesizing and evaluating are associated with productive modeling (Löhner, van Joolingen, Savelsbergh, & van Hout-Wolters, 2005). Therefore, the experiments students have to perform in this lesson design are scaffolded with a worksheet on which they have to write down hypotheses and evaluations. The teacher/researcher will encourage students to use this worksheet, preventing that students use the modeling software as a game or puzzle instead of a scientific inquiry tool. Even with the final modeling task (modeling an action potential), which requires more student input, students are asked to first write down the ion currents that permit this change of membrane potential and the proteins that facilitate the flow of ions before they start working on the computer model. These measures to scaffold meaningful inquiry learning overlap with the ways in which students are encouraged to reason mechanically because both inquiry and mechanistic reasoning are authentic scientific skills that students need to successfully construct and explain their models.

Provoking mechanistic reasoning

During the modeling task students are encouraged to use mechanistic reasoning techniques, thereby teaching them to explain the system meaningfully. We aim to stimulate mechanistic reasoning without mentioning it explicitly. This is done in two ways. The first assignments on the worksheets are designed such that students need to use specific elements of mechanistic reasoning in order to complete the assignment. This is the scaffolding phase in which students practice with reasoning skills, activate prior knowledge and learn to use the software. Furthermore, the teacher/researcher demonstrates elements of mechanistic reasoning and asks questions to stimulate students to verbalize mechanistic explanations.

In order to approach the provoking and measuring of mechanistic reasoning systematically, a list of elements of mechanistic reasoning was composed for this research based on the coding scheme of Russ et al. (2008). The scale is adapted for the present research in the following way: 'Describing the Target Phenomenon' makes place for 'Asking how-questions'. Furthermore, an extra category (-1) is added to mark reasoning that is the opposite of mechanistic reasoning, Teleological or anthropomorphic statements (TS). The actual coding scheme is found in Appendix C. In the following paragraph it is explained how these elements of mechanistic reasoning will be stimulated during the assignment:

1. Asking how-questions: This element is weakly stimulated by the first assignment because it is an example: students have to write down *how* a neuron manages its resting potential. For this element the teacher has a more important role: during the whole modeling task the teacher reacts on statements of students by asking how it works. For example, when a student hypothesizes that by opening a sodium channel the neurons membrane potential will become negative, the teacher asks how this works, forcing the student to propose a mechanistic explanation. Students might learn from this behavior and ask their partner for mechanistic explanations during the modeling tasks.
2. Identifying Setup Conditions, Identifying Entities, Identifying Activities, Identifying Properties of Entities and Organization of Entities: application of these elements are all practiced during the informal model drawing task. Students are asked to write down structures and their functions and subsequently drawing a model to represent the activities and organization. Moreover, during the first guided experiment, students are asked to write down properties of ions.
3. Chaining: Chaining is stimulated every time students are asked to write down hypotheses (Experiment 2, 3, 4 and 5). In the fifth experiment students get a challenging assignment: hypothesizing how to make a negative membrane potential only by adjusting the membrane channels. Students are asked first to explain which adaptation they think that will lead to the desired result, thereby forcing them to use chaining.

Collaborative reasoning

During the assignment students work in dyads. This setup was chosen in order to tap students' reasoning through their conversation (Teasley, 1997). Furthermore, several studies and widely accepted educational theories indicate that effective collaboration yields higher learning outcomes and task performance (Samaha & De Lisi, 2000; Rojas-Drummond & Mercer, 2003).The benefits even go further than the task which is done collaboratively, because talking with a peer while working on a task also enhances development of scientific reasoning skills and understanding of the study phenomenon (Mercer, Dawes, Wegerif, & Sams, 2004).

Research design

The following scheme presents an overview of the hypotheses that will be tested in this study, expected evidence that confirms these hypotheses and the associated way of data collection.

Table 1

Relationship between hypotheses and data collection for this study

Hypothesis	Evidence	Data collection
It is possible to teach students to use mechanistic reasoning during a guided modeling task.	During the task students gradually show more and higher order elements of mechanistic reasoning. Students use higher order elements of mechanistic reasoning in the posttest question.	Score of verbal reasoning on audio → focus on verbal reasoning during 'free' modeling task because reasoning is not scaffolded during this task. Score of reasoning on worksheets
Application of mechanistic reasoning leads to better performance on modeling tasks	Students who use more and higher-order elements of mechanistic reasoning during a task perform better (sophisticated and complete model, use of prior knowledge, underpinning adaptations, use less time, less trial-and-error) and show more insight in the process of modeling in the 'reflection on modeling' assignment.	Comparison of reasoning scores during a modeling task and modeling performance SimSketch logs Score of assignments that reflect on the modeling task
A learning trajectory in which a computer modeling is combined with stimulation of mechanistic reasoning is an effective method for learning complex biological systems	The final model score is significantly higher than the pretest hand-drawn model.	Pre- and posttest model
	Students show deep insight in the posttest question.	Score posttest question
	Students verbalize deep insight in the system during the modeling tasks.	Audio recording

Method

Participants

Eight preuniversity students in the final year of the Dutch secondary school education participated in this study. All students followed biology classes. During the two-hour experiment they worked most of the time in couples. Dyads were formed by letting the students choose a partner. To ensure anonymity we replaced their real names with pseudonyms generated using http://www.behindthename.com/random/

Instruments

SimSketch

In the SimSketch application designed for modeling a functional neuron, students are able to perform experiments with a simulated membrane (Bollen & van Joolingen, 2013). All objects except the membrane itself are drawn by the students. After drawing, tags can be added to drawn objects to make the drawing functional. SimSketch measures the transmembrane voltage, so voltage-gated ion channels will respond to the voltage. Also, the transmembrane voltage is shown in a diagram. Thresholds for voltage-gated ion-channels are set by the student, as are the location and number of all previously mentioned items. Thereby, students can build their own model of a neuron, experiment with it, evaluate the outcomes and revise their model.

Assignments

The guided experiments are loosely based on a series of experiments described in Neuroscience (Purves, et al., 2008), a widely used textbook in academic life sciences studies. The final assignment for measuring deep insight is based on a question in a Dutch biology exam. The assignments are included in appendix A.

Data collection

During the lesson, the following data was collected: Individual prior knowledge, score of informal model of dyad, reasoning style on worksheets (modelling assignments), vocal reasoning style of dyads, SimSketch Logs, SimSketch model score, post tests (insight in modelling process, question on deep insight in phenomenon). The scoring system is included in appendix B.

Analysis

Analyses of the reasoning processes and modelling actions of students during the action potential assignment were performed by aligning students' conversations, SimSketch logs and worksheets. These were analyzed in order to find episodes of mechanistic reasoning to investigate whether mechanistic reasoning was effective. Episodes of reasoning that led to modelling actions were analyzed in order to find out whether students did reason mechanically in these episodes and if the reasoning action did contribute to the modelling process. During the analysis of the data it became clear that students were using the lower and mid-order elements like identifying entities and their properties and activities all the time and it has thus no discriminative value to score these elements. Therefore, it was chosen to name a reasoning fragment higher-order mechanistic reasoning only when there was evidence of 'identifying organization' or 'chaining'. At last, evidence for learning and deep understanding was looked for to find out if the students actually gained deeper understanding of the phenomenon. The full analysis can be found in the appendix 'Analysis of reasoning and modelling actions'.

Results and discussion

Pilot test (pilot)

The learning material was tested with one participant, a nurse in training, in a pilot study in order to fine-tune the assignments and software.

First Test

In this test four 6 VWO students (two male, two female) participated and formed dyads of their own choice.

Prior knowledge

The four students had very little prior knowledge about neural regulation and none of them was able to explain resting potential correctly in the individual pretest. Both groups only drew a sodium-potassium pump in their informal model with no channels and no other particles than sodium and potassium.

Provoking mechanistic reasoning during the experiments

There are clear signs of mechanistic reasoning during these episodes of hypothesizing and when the students were running the model to check if their hypotheses were right. Leon and Veer used mechanistic reasoning but often formulated an incorrect hypothesis. This might be due to their lack of knowledge about charged particles. However, they quickly learned about this phenomenon, because in the third experiment they successfully predicted the effect of a sodium channel on the membrane potential:

Leon: *Because that one goes over here, so now it will get more positive because positive ions move into it.*

Afterwards they discussed what exactly caused the change in membrane potential and formulated a mechanistic explanation for the change in potential together:

Leon: *The positive sodium ions will go inside, right?*
Veer: *Yes, but look, the negative ions will stay behind.*

Stijn and Isabelle made better predictions than the other group, and based their hypotheses on higher-order mechanistic reasoning steps, like in this fragment of hypothesizing during experiment 2:

Stijn: *So now potassium ions can go outside, so inside it will get more negative, so I think the potential gets more negative.*
Isabelle: *Yeah I think so.*
Stijn: *Because positive ions can go outside now.*
Isabelle: *But they cannot go inside.*
Stijn: *They can also go inside.*
Isabelle: *Yes the potassium things, but not the sodium things.*
Stijn: *Exactly.*
Isabelle: *So at first it will definitely will become more negative.*

Reasoning and modeling during the action potential assignment

For the first part of this assignment, the students have to write down which ion flow and ion channels facilitate the different stages of an action potential. They are given a graph that represents the change in membrane potential during an action potential. Both groups discussed how ions and ion channels facilitate action potentials and both groups use mechanistic reasoning, but the proposed mechanisms are completely different. Veer and Leon came up with a mechanism that could be theoretically correct, but differs from reality. They tried to model this only with clickable channels, so they could manually simulate an action potential. It is not completely clear why they did not use voltage-gated channels in their first model, but from the following fragment it seems that they just did not knew how to make these channels:

Leon: *I want to trigger them all by mouse clicks.*
Veer: *Why?*
Leon: *So you can regulate them and you can open this one so it goes to the threshold value and then open this one so it goes up and then close them all and open this one so it goes down again.*
Researcher: *But what is the second step?*
Leon: *Oh right, that is with the voltage-gated things. But how can we make those?*

In the other group, Stijn starts the discussion by explaining what happens during depolarization: suddenly positively charged sodium ions flow into the cell and thereby making the inside of the cell more positively charged. According to him, the sodium-potassium pump is the only active membrane protein during resting potential, which is in line with the informal model of this dyad. Eventually, they described the ion currents correctly, but they did not mention different types of channels or what triggers opening or closing of the channels.

Both groups needed encouragement from the researcher before they started talking about what type of ion channels (ligand-gated, voltage-gated and always open) were needed for the different steps. Moreover, both groups at first made the same mistake when they talked about the threshold value for opening the sodium channel. They just copied the threshold value (-50mV) from the graph without taking into account what is possible with their own model. However, they created models that were in line with their initial reasoning and by making and running these models, they figured out that they were wrong and started to reason again. Eventually, both groups were able to create a model in which the steps of an action potential can be simulated and both groups needed quite a lot of support from the researcher to get there. However, there are some remarkable differences. Stijn and Isabelle were quicker than the other group, they used less steps to get all the necessary ion channels in place and had enough time to run the model for several times and fine tune channel settings. Their model had a higher score (8,5) than the other group (6,9). Veer and Leon were slower and had fewer cycles of running and improving the model. They just had all the necessary membrane proteins before they ran out of time.

Posttests

Insight in modelling process

All students could think of a meaningful addition to the modelling software, for example the ability to add neurotransmitters, ATP, and the possibility to work with more ions. Stijn and Isabelle both briefly described a research aim for an experiment that could be performed with a virtual membrane. Stijn: How mineral deficiency influences impulse transmission. Isabelle: How drugs influence the functionality of ion channels. Veer and Leon did not come up with meaningful experiments.

Deep insight question

Stijn was the only student who answered correctly and explained his answer properly using chaining, the most sophisticated expression of mechanistic reasoning:

As a result of sleeping pills, one becomes less active so less impulses are taking place. When Cl^- ions are able to enter the cell, this counteracts impulse transmission because this influx hyperpolarizes the membrane. So I think that Cl^- channels open more easily due to the sleeping pills.

The other students did not give correct answers, but Leon and Veer did use chaining. So they might have learned how to explain a phenomenon mechanically but were still not familiar enough with this subject to work with it.

Conclusions from this round

- The students worked enthusiastically for two hours on the assignments. They were able to finish all assignments although they needed extensive help from the researcher.
- There are signs of mechanistic reasoning in students' conversations during the scaffolding phase. This happens mostly when they are hypothesizing about the outcome of an experiment and when discussing results.
- During the action potential modeling task there is evidence of mechanistic reasoning, but this occurred mostly during the first phase when the students are asked to describe which ion currents an ion channels facilitate the various stages of an action potential.
- The modeling actions are consistent with the students' initial discussion of action potential.
- Only one student answered the final question correctly, but three out of four students used sophisticated mechanistic reasoning in their answers.

Implications and modifications for the next round

It was clear that all the students had very little prior knowledge of the molecular mechanism that enables nerve cells to maintain their resting potential. Even the best performing student mentioned only the sodium-potassium pump when asked to describe how the resting potential is maintained. Moreover, the final insight-question seemed too difficult for 3 of the 4 students. This lack of prior knowledge was distracting the students from reasoning about the mechanics. Therefore, it was decided to provide the students with some extra information about the workings of the nerve cell membrane. This information consists of visual models of the resting membrane potential and action potential, taken from BINAS, a reference book which students are allowed to bring on the Dutch national exams for biology. This is handed out after the prior knowledge test, the researcher then explains that with SimSketch we model a membrane of a nerve cell and uses the model in BINAS to explain what this is. This modification has another advantage: Students now use SimSketch to bring the information in BINAS to life. In this way, students are able to create a dynamic model based on the static model they are already familiar with. Expectantly, this allows students to connect their prior knowledge better to the assignment with SimSketch.

The modelling assignment was modified by adding a new step to make clear to the students that the model they made in SimSketch describes the same phenomenon as the model they know from BINAS. Therefore, the students first compare their model, derived from experiment 5, with the model of the resting potential from BINAS. Then they describe how their model differs from the model in BINAS. Because it seemed too difficult for students to describe ion flows and the activity of ion channels during action potential, and because it took the students a lot of time to get clear that there are different triggers for opening ion channels, in the new version the students have to study a complete model of the action potential from BINAS in which the ion channels are visualized for every phase of the action potential. They have to discuss which ion channels facilitate the ion currents and how this influences membrane potential. Then they describe how to model this in SimSketch and they are asked to think about what trigger opens the ion channel for every step.

The instruction for the modelling assignment was also modified. In the previous version students were told to adapt their model derived from experiment 5 to create an action potential. A list of tips were given for a quick start. Students read some of the tips, but not all, and it seemed that this distracted them from creative thinking of how to make a better model. Consequently, in the new version students are told they can use experiment 5 as a starting point and that they are free to choose what they keep and what they delete. In order to provoke more discussion and hopefully more signs of mechanistic reasoning, only two additional instructions are given by the modeling task (instead of a list of tips): Every time you run the model you discuss what you see and how you are going to adapt the model, and: change only one thing and then run the model to evaluate the effect of the adaptation. To make it more clear for students that there are three different types of ion channels, the extra icon for a clickable channel is deleted and this function is integrated in the 'open when' drop down menu.

Second Test

In this test four 6 VWO students (all male) participated and formed dyads of their own choice.

Prior knowledge

These students had slightly more prior knowledge than the students in the previous test. Ewout and Karel drew not only a sodium-potassium pump but also ion channels. Arie and Ruben drew a sodium-potassium pump with the correct number of exchanged ions and were the only dyad that included chloride ions in their informal model.

Provoking mechanistic reasoning

All students showed elements of mechanistic reasoning and the mechanistic reasoning episodes happened roughly at the same moments as in the previous test: when making predictions and when running the model. In this test, a beautiful example of how specific language can enhance the reasoning process was observed. During the second experiment, Ewout introduces the word 'spreading' to describe the movement of ions when they are able to pass the membrane, which seems to help the students to reason mechanically about the behavior of the ions.

Ewout: *So it becomes more negative, or could this reverse also? No, even then it should be more negative because it spreads.*
Karel: *Oh right, potassium will spread so…*
Ewout: *…on the outside there will be a higher voltage.*

They keep using the concept of spreading to make predictions in experiment 3:

Ewout: *Yes that is the same, it spreads over the two, the sodium. Yes it is the same but reversed, sodium spreads so here it will become higher.*

Arie and Ruben often were in disagreement, so they argued a lot. This more often results in episodes of mechanistic reasoning, but it also slows down the reasoning process because sometimes they just do not listen to each other's arguments. This is an example of how social interactions can distort instead of enhance reasoning when students are working in dyads. Arie proposes a strategy to lower the potential, but Ruben overruled this with another (less effective) strategy. Another thing that is interesting about the following fragment is that again the students overestimate the effect of the sodium-potassium pump, which is observed several times during this research.

Arie: *We want it to become more negative, so we want more potassium to leave.*
Ruben: *Yes, just make the channel as small as possible.*

Arie: *So the sodium channel has to be very small.*
Ruben: *Both of them have to be very small.*
Arie: *But then it does not become negative because it cannot leave.*
Ruben: *When they are very small then the pump does everything.*
Arie: *All right then.*

Reasoning and modeling during the action potential assignment

The dyad Arie and Ruben did not cooperate efficiently during this part of the assignment. They use only incomplete sentences and did not show higher order mechanistic reasoning such as chaining. After the initial discussion they wrote on the worksheet: *The channels have to open when the voltage gets higher than the threshold value.*

Ruben takes the lead in these fragments and at first he gives two brief explanations of the mechanism of action potential: 1. It (the sodium channel) is closed until threshold is reached (and then they open exceedingly). 2. They (the sodium channels) open when they are sufficiently stimulated. When the students have read the assignment fully and have learned about the three types of ion channels, Ruben start anew with reasoning: When both channels are closed, nothing happens, the channel has to open when the threshold of -50 mV is reached. He seems not so sure if the channels must open when the thing (membrane potential) gets higher or lower than the threshold. Their final description of the mechanism is incomplete and only focusses on how depolarization takes place, neglecting how the threshold is reached and how the membrane repolarizes.

The other group has a completely different discussion, they cooperate and use chaining in order to explain the mechanism:

Ewout: *As a result of the threshold the sodium channels open.*
Karel: *And then this one becomes really high. And then in the inside it becomes really high. And during depolarisation it becomes even higher, so during repolarization it becomes lower.*
Ewout: *Yes so during a certain high voltage these have to come open.*
Karel: *Yes, that is the potassium.*
Ewout: *No, that is during a very low voltage. No sorry at a high voltage.*
Karel: *And at the threshold value sodium channels. I think it is correct like this.*
Ewout*: and the pump takes care of this?*
Karel: *Yes.*

They wrote on the worksheet:
K- and Na-channels.
At a very high voltage, K-channels open.
At threshold, Na-channels open.

So the mechanism suggested by Ewout and Karel is very close to the textbook explanation: Only the potassium and sodium channels play a role in this process (not the sodium-potassium pump). At threshold, sodium channels open with a high potential (more positive) as a result. During depolarization, it gets even higher. During repolarization, K-channels open because of a high (more positive) voltage. Their explanation lacks a clear difference in the difference between a small change in membrane potential (from resting potential to threshold potential) and the actual depolarization.

The different qualities of the reasoning processes of these two groups are also found in the way they work on the model. Ewout and Karel relatively quickly drew and set all necessary channels and had

time to fine-tune their model. Their model was the best representation of an action potential and got 10 out of 10 points according to the scoring system.
Arie and Ruben, in contrary, lost a lot of time by clicking around in the software, checking out channel functions etc. They needed external motivation from the researcher, who pointed out the different stages of action potential, to start reasoning about the mechanism again. Then they came up with a more complete mechanism, however their reasoning is still inaccurate (they talk about particles getting more positive) and there is no chaining:

Ruben: *Okay, Phase one is closed.*
Arie: *First only sodium goes. Only potassium is closed.*
Ruben: *The stimulus will close the channels, that is a mouse click. And when the cannels open, sodium is green, so then the particle will become more positive. So then, when it gets even more positive then have to open more, or something?*
Arie: *Yes more of them will open.*
Ruben: *So you need two channels or so.*

It is notable that these students really learn something from working with SimSketch. They do not have a well-defined internal model for this phenomenon, but during the modelling assignment they discover the gap in their initial proposed mechanism, they forgot to think about how hyperpolarization takes place:

Arie: *But how does hyperpolarization takes place?*
Ruben: *Because way more particles will go out than go in. Because of the pump.*
Arie: *So the pump it restores after a while, right?*
(Students are observing the model)
Ruben: *Why doesn't it make it sink back?*

Initially they think that the sodium-potassium pump will repolarize the membrane. However, in the last minutes of the assignment, there is proof that at least Ruben understands that the influx of potassium is the mechanism for repolarization:

Arie: *So those will endlessly go open, the potassium?*
Ruben: *No, those quit eventually, when it's hyperpolarized again.*

Their final model contains the necessary ion channels to model an action potential. However, the voltage-gated sodium channel is smaller than the clickable channel, and the threshold of the potassium channel is too high to reach with this model.

There is also evidence that the students of the more successful group also learned during the modelling process. They created a model with two voltage-gated channels, one for sodium and one for potassium, but did not create a channel that could deliver the initial stimulus for the action potential. So their modelling actions were in line with their reasoning, which resulted in a model in which nothing happened:

Ewout: *It doesn't do anything, no it I stuck at -2.*
Karel: *Yes but it should change soon. And then it should become -5.*

The students were stuck at this point, so the researcher helps them remember that there are three different types of channels (always open, clickable, voltage-gated). Karel seems to understand that they need another sodium channel in order to facilitate the two different influxes of sodium ions:

Karel: *Okay look we set this trigger on 'greater than' and then just that one. We make another one that opens on a mouse click.*

After drawing the new channel, the students became enthusiastic because the model behaves like they want to: in the audio record Ewout says*: 'And now we have some sort of a peak, and then it goes like this, and now...'* followed by yelling and clapping. Subsequently, they ran the model several times and adapted channel settings quickly, resulting in the best model created during this study, as shown in figure 1.

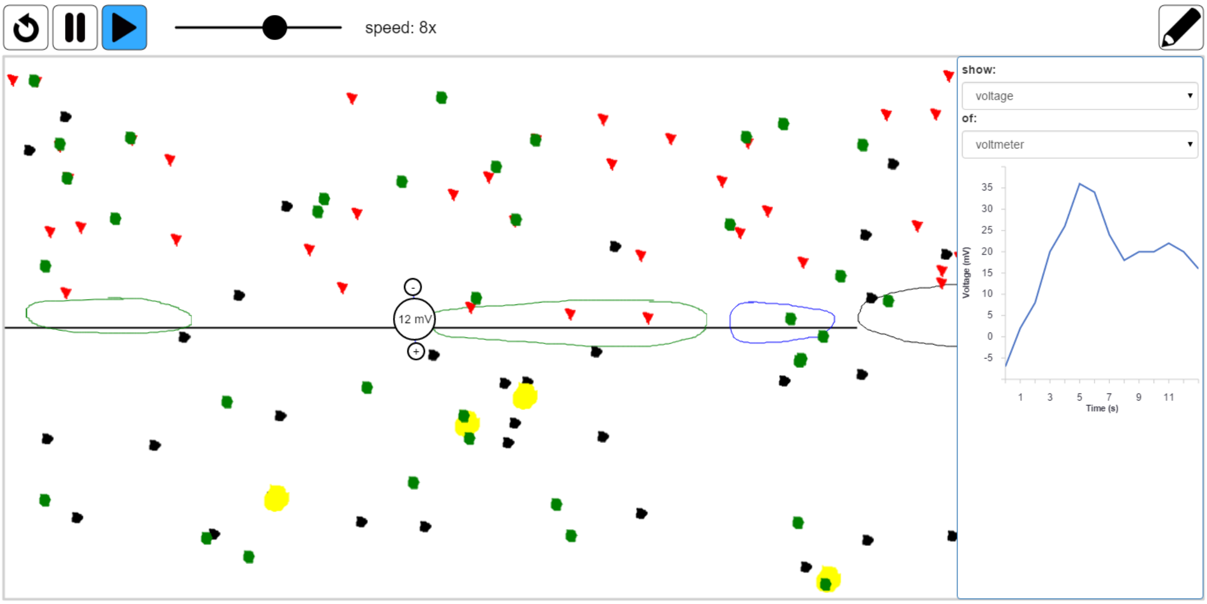

Figure 1. Model representing action potential created by Ewout and Ruben. The open shapes on the horizontal black line represent membrane proteins and the smaller filled shapes represent charged particles. A clear peak is visible in the graph on the right, representing the change of electric potential during the simulated action potential in this model.

Posttests

Insight in modelling process

Again, all students proposed meaningful additions to the software, like neurotransmitters, more ions and a longer piece of membrane to simulate how an action potential propagates. Arie and Ruben did not describe a meaningful experiment that could be performed with a virtual membrane. Ewout and Karel both only briefly described an experiment in which is studied what happens when a neuron gets a lot of stimuli in a short time span.

Deep insight question

Contrary to the previous round, students could use a picture from BINAS during this assignment that showed the concentration of ions inside and outside the neuron. 3 out of 4 students came to the correct answer. Two of them, Ruben and Karel, also showed the proper mechanistic reasoning steps that have led to this answer. Karels answer is a nice example of the reasoning style that was aimed for:

- *When the potential is lower, stimuli are less likely to be transmitted.*
- *Assuming that less stimuli results in sleep,*
- *The potential has to be lowered by the sleeping pill.*

- *Cl- is negatively charged, so the Cl- channels have to open more easily.*

Conclusions from this round

- Providing a visual model from BINAS during the initial discussion about the mechanics of an action potential led in one group to a meaningful and mechanistic explanation of the phenomenon that was near a textbook explanation. This dyad also created the best model.
- In the other group, however, this was not the case. They did not show higher-order elements of mechanistic reasoning during the discussion of action potential and their final model was less complete.
- Providing a visual model from BINAS during the posttest deep insight question seems to have enabled students to get to reason mechanically which led to the correct answer.
- All students showed to be able to reason mechanically, especially when they were asked to make predictions and to test their predictions.
- The group that showed higher order elements of mechanistic reasoning during the discussion about action potential also made the best model.
- In both groups there is proof that they learned by interaction with the model.
- All students were enthusiastic about the assignment and the software.

Conclusions and implementations for teachers and designers

From the pretest data it is clear that the participating students were not able to fully explain resting potential using a mechanistic model at the start of the lesson. The best explanations only mentioned the sodium-potassium pump but these were in no way mechanistic explanations. This is a sign that these students have not yet learned to understand complex biological systems thoroughly and supports the quoted literature on difficulties with complex biological systems in secondary school biology classes.

It is concluded that it is possible to teach students to use mechanistic reasoning using a computer modeling assignment. Asking them to make predictions and to evaluate these predictions with modelling software led to mechanistic explanations. The assignments must be scaffolded, so students are forced to think before they act, and evaluate their reasoning afterwards. This alternation between reasoning and modelling is not only a powerful way to stimulate reasoning, it also helps students to efficiently choose reasoning actions. Dyads that showed more convincingly that they were able to reason mechanically also performed better on the final modelling task and this was also reflected in their answers on the final deep insight question. Although this assignment requested hard work and costs two hours, the participants barely complained and worked enthusiastically, were most of the time on-task and reported to think this assignment is both fun and educative. This might be (except for the fun of drawing your own model and just doing something else than regular classes) due to the fact that during biology classes there is usually no time to go deep and really understand a subject, and in this assignment the students gained deep understanding which is very rewarding.

For teachers and educational designers this means that they must be aware it is possible and very rewarding to guide the reasoning processes of students. This research has shown a computer modelling task is an environment that makes this easily accessible because of the possibilities to hypothesize and experiment. Although it is sometimes difficult because of the overcrowded curriculum to go deeper into a subject to unravel the mechanism, students really appreciate it and are willing to work for it. So I would advise to take the reasoning abilities of preuniversity students seriously and to create interesting assignments that challenge them to open the black box and to exceed just remembering concepts.

Limitations and further research

The current research was small scaled and explorative of nature. It provided the first evidence that combining computer modelling and provoking mechanistic reasoning is an efficient way of gaining understanding of complex systems, and it has its limitations. There was a lot of interaction between the researcher and the participants, making it more difficult to say whether successful reasoning or modelling steps are the result of the composition of the assignment and software. Moreover, the qualitative analysis made this research dependent on the choice of the author to implement or interpret data. Therefore, further research on this topic should be done on a larger scale and should leave less space for the personal choice of the researcher to interpret data and to interfere with the participants. This could be done by adapting the software and the worksheets so the software itself provides feedback and guidance for the students and test it in a classroom setting. This way also quantitative data could be collected to supplement the findings of the current research. Also, it is interesting to test this technique of guiding reasoning during a computer modelling task in other science domains to study the generalizability of these findings.

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Appendix A: Assignments

Voorkennisopdracht

Deze opdracht maak je individueel:

Zenuwcellen hebben in rust altijd een bepaalde lading, dit heet de rustpotentiaal. Beschrijf zo precies mogelijk hoe een zenuwcel dit rustpotentiaal in stand houdt.

Gezamenlijk informeel model tekenen

Deze opdracht maken jullie in duo's.

1. Schrijf in onderstaand vak op welke celstructuren, moleculen en ionen een rol spelen bij het reguleren van de membraanpotentiaal.
2. Beschrijf ook de functies en eigenschappen van elk onderdeel.
3. Maak op de volgende pagina een tekening waarin je weergeeft hoe alle onderdelen georganiseerd zijn tijdens een rustpotentiaal.
4. Geef de stroom van de ionen aan met pijlen.

SimSketch opdrachten

Deze opdrachten maken jullie in duo's

Experimenten uitvoeren met SimSketch

We gaan een serie experimenten uitvoeren met een virtueel membraan. Door middel van deze experimenten leer je het programma kennen en doe je kennis op over de werking van het membraan van zenuwcellen.

Experiment 1

We gaan stap voor stap het eerste experiment klaarzetten. In dit experiment hebben we aan de binnenzijde van het membraan 30 kaliumionen en 3 grotere negatieve deeltjes. Aan de buitenzijde zijn 30 natriumionen en 30 chlorideionen. Vergeet niet objecten te groeperen met de lasso als je een tekening maakt met meerdere lijnen.

1. Teken aan de binnenzijde van de cel een kaliumion en een eiwit
    a. Geef beide deeltjes een andere kleur
    b. Benoem de deeltjes
    c. Voeg nu de juiste lading toe door een + of – op de ionen te slepen, geef kalium een lading van +1 en het eiwit een lading van -10, geef het eiwit ook een grotere massa
    d. Sleep een fabriekje naar de ionen en stel de juiste aantallen in
2. Teken aan de buitenzijde van het membraan een natriumion en een chlorideion
    a. Geef beide deeltjes een andere kleur
    b. Benoem de deeltjes
    c. Voeg nu de juiste lading toe door een + of – op de ionen te slepen, geef natrium een lading van +1 en chloride een lading van -1)
    d. Sleep een fabriekje naar de ionen en stel de juiste aantallen in
3. Teken een voltmeter als een draadje tussen de twee compartimenten
    a. Een voltage is het verschil in lading tussen het ene en het andere compartiment, de richting waarin je tekent bepaalt welk compartiment je meet (begin van het draadje) en welke de referentie is (einde van het draadje)
    b. Benoem het object
    c. Sleep het icoon met de voltmeter op het object
4. Run nu het model
5. Observeer hoe de ionen bewegen. Welke wetmatigheden bepalen de beweging van de ionen?

Experiment 2

1. Teken in het membraan een kaliumpoort die altijd open is
    a. Teken de poort (niet te klein!) in het membraan met dezelfde kleur als de kaliumionen
    b. Sleep het poort-icoontje op de poort en kies de juiste functies
2. Hoe denk je dat het membraanpotentiaal verandert gedurende de tijd? Streep door wat niet van toepassing is
    a. Het potentiaal: wordt negatiever / blijft gelijk / wordt positiever
    b. Beredeneer je antwoord

3. Run het model, klik op de grafiek en kies ‘voltage’ en ‘voltmeter’ om de veranderingen in het voltage te volgen.
4. Bespreek wat je ziet.
5. Klopt dit met je verwachting? Zo niet, beschrijf hoe dit verschilde met wat je dacht.

Experiment 3

1. Vervang de kaliumpoort door een natriumpoort.
2. Hoe denk je dat het membraanpotentiaal verandert gedurende de tijd? Streep door wat niet van toepassing is
   a. Het potentiaal: wordt negatiever / blijft gelijk / wordt positiever
   b. Beredeneer je antwoord

3. Run het model.
4. Bespreek wat je ziet.
5. Klopt dit met je verwachting? Zo niet, beschrijf hoe dit verschilde met wat je dacht.

Experiment 4

Membranen in zenuwcellen zijn altijd een beetje lek voor zowel natrium als kalium. De natrium-kaliumpomp houdt de verschillende concentraties in stand door drie natriumionen naar buiten te pompen en tegelijkertijd twee kaliumionen naar binnen.

1. Houd de ionconcentraties hetzelfde
2. Maak zowel een kaliumpoort als een natriumpoort van ongeveer dezelfde breedte
3. Teken een natrium-kaliumpomp en stel deze juist in door middel van het icoontje met de pomp
4. Hoe denk je dat het membraanpotentiaal verandert gedurende de tijd? Streep door wat niet van toepassing is
    a. Het potentiaal: wordt negatiever / blijft gelijk / wordt positiever
    b. Beredeneer je antwoord

5. Run het model voor een wat langere tijd.
6. Klopt je observatie bij wat je verwachtte? Zo niet, beschrijf hoe dit verschilde met wat je dacht

Experiment 5

1. Een zenuwcel in rust heeft een negatief membraanpotentiaal. De cel regelt dit onder andere door sommige ionen gemakkelijker over het membraan te laten verplaatsen dan anderen. Probeer daarom het potentiaal negatiever te maken door alleen de doorlaatbaarheid voor kalium en/of natrium te veranderen. Dit is het makkelijkst door de poorten groter of kleiner te maken.
2. Beschrijf eerst wat je wilt aanpassen en beredeneer waarom je denkt dat dit het potentiaal negatiever maakt.

3. Probeer uit of de aanpassing het gewenste effect heeft, blijf experimenteren totdat je een overtuigend negatief geladen cel hebt gemaakt. Welk ion moet het meest doorgelaten worden en waarom zorgt dit voor een negatief potentiaal?

4. We hebben nu de doorlaatbaarheid van het membraan veranderd door het formaat van een poort te veranderen omdat dit gemakkelijk en overzichtelijk is in het model. Hoe denk je dat dit in een echte zenuwcel geregeld is?

Vergelijking model SimSketch en model BINAS

Jullie hebben nu een model gemaakt in SimSketch waarmee verklaard kan worden hoe een zenuwcel zijn rustpotentiaal behoudt. Als het goed is lijkt dit model op het plaatje in BINAS (Zie infoblad, rustpotentiaal) waarin het rustpotentiaal wordt weergegeven. Bespreek met elkaar in hoeverre jullie model overeen komt met het model in de BINAS en beschrijf één verandering die jullie model nog nauwkeuriger zou maken.

Actiepotentiaal

Actiepotentiaal in de BINAS

In het plaatje uit BINAS is samengevat hoe een actiepotentiaal verloopt. Bespreek samen aan de hand van de plaatjes hoe het openen en sluiten van de poorten het membraanpotentiaal (weergegeven in grafiek) beïnvloedt.

Actiepotentiaal modelleren in SimSketch

In SimSketch kan je poorten op drie verschillende manieren instellen: altijd open, open bij bepaald voltage en open op muisklik. Schrijf in het kort op welke poorten je nodig hebt om een actiepotentiaal te kunnen modelleren. Beschrijf niet alleen welk ion de poort moet doorlaten maar ook op welke trigger de poort moet openen.

Opdracht: modelleer een actiepotentiaal in SimSketch

Gebruik nu SimSketch om een model van het actiepotentiaal te maken waarin je elke fase van het actiepotentiaal kan doorlopen. Gebruik het model van opdracht 5 als startpunt, je mag zelf weten welke onderdelen je van het model van experiment 5 bewaart en welke je verwijdert/verandert.

Bespreek altijd met elkaar wat je ziet als je het model runt en hoe je het model gaat verbeteren. Verander dan altijd maar één ding voordat je het model opnieuw runt zodat je weet wat het effect is van deze verandering.

Reflectie op het model en modelleren

Deze opdracht maak je individueel

Een membraan van een zenuwcel zit in het echt natuurlijk veel ingewikkelder in elkaar dan wat je kan laten zien in dit model. Noem twee aanpassingen die het model meer waarheidsgetrouw zouden maken.

Wetenschappers gebruiken ook modellen om biologische systemen te verklaren of om voorspellingen te doen. Beschrijf een wetenschappelijk experiment die met een virtueel membraan uitgevoerd zou kunnen worden.

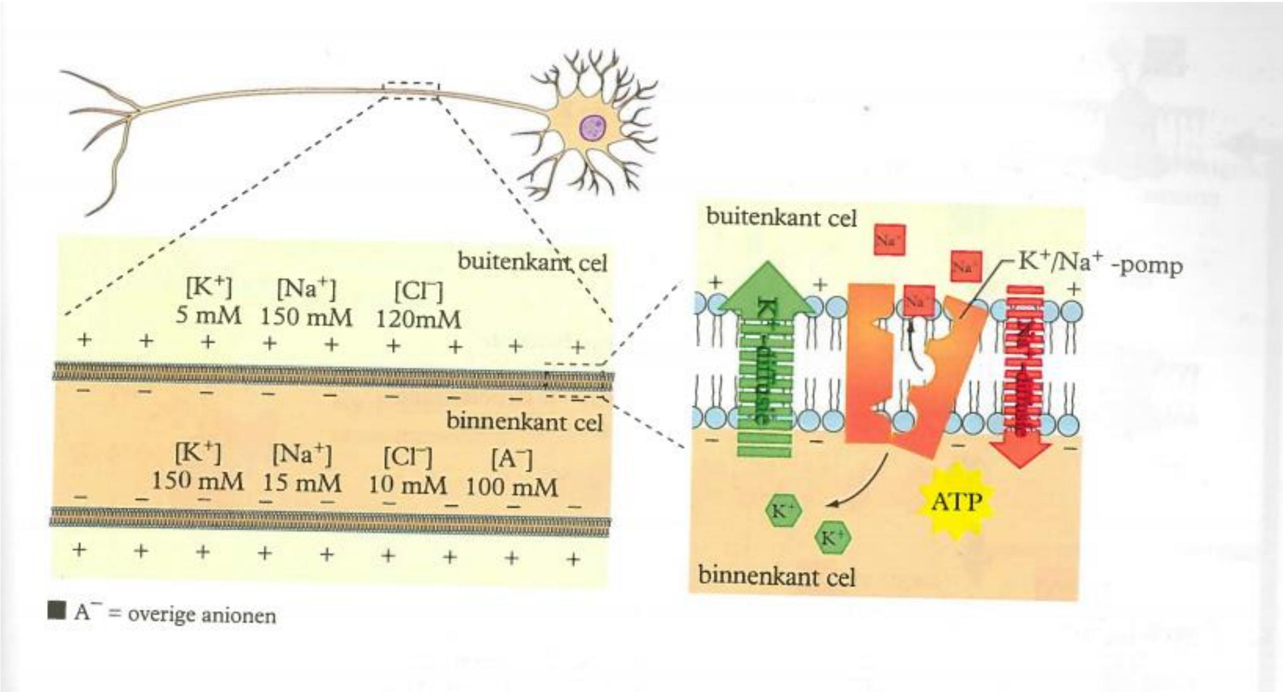
Eindopdracht

Deze opdracht maak je individueel

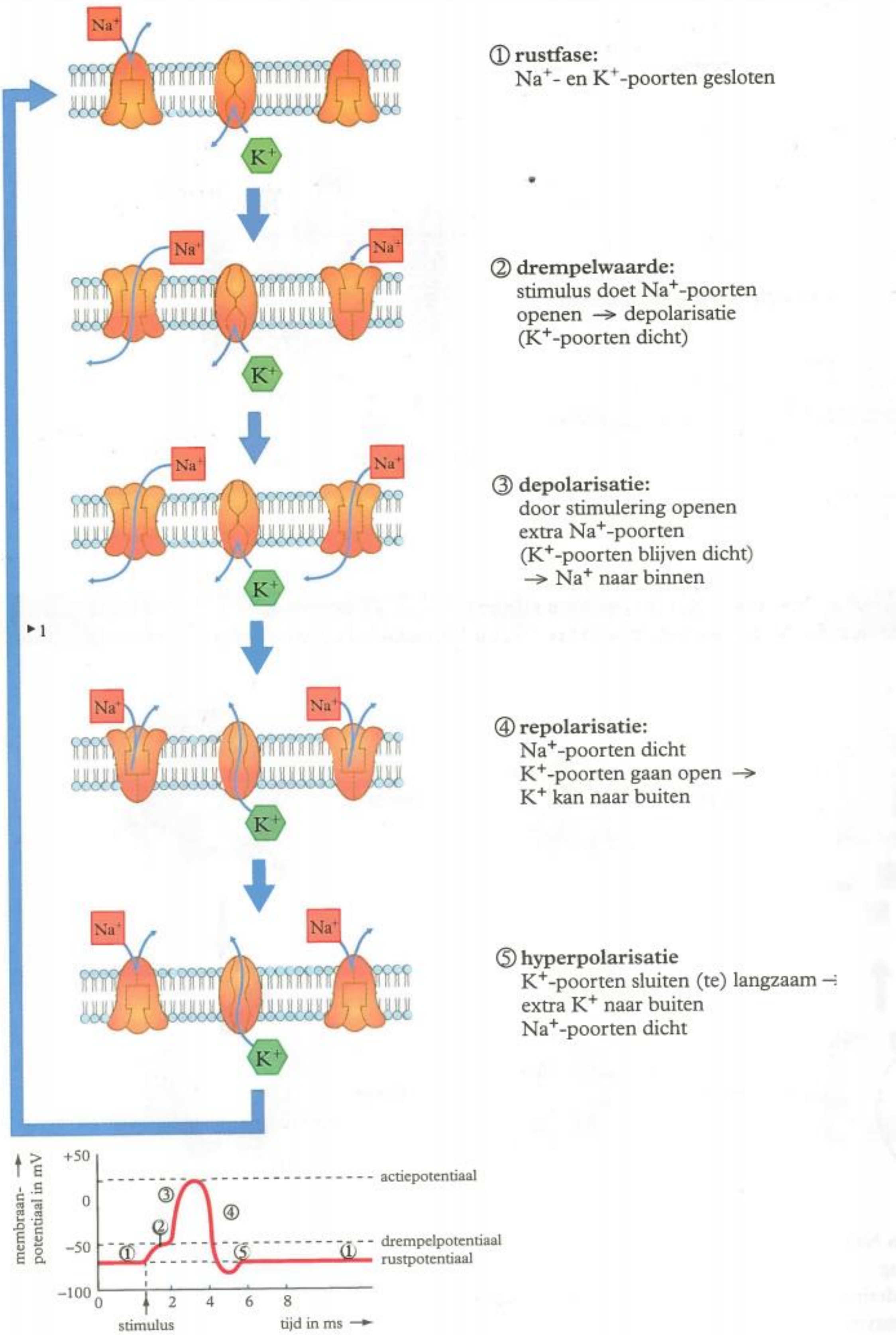
1. Er zijn stoffen die de werking van de chloridepoorten beïnvloeden, zoals slaapmiddelen. Denk je dat door een slaapmiddel deze poort juist gemakkelijker of moeilijker opent? Beredeneer je antwoord zo volledig mogelijk.

Infoblad

Rustpotentiaal



Actiepotentiaal



Appendix B: Scoring of assignments

Scoring hand-drawn model (20 points in total)

Divide the total score by 2 for grading

Structures:

The model contains the following structures (1 point for each structure, 8 points in total):

- Potassium ions
- Sodium ions
- Chloride ions
- Negatively charged proteins / RNA
- Membrane
- Potassium channel
- Sodium channel
- Sodium-potassium pump

The model is correctly organized (6 points in total)

- Extracellular concentrations of sodium and chloride are higher than intracellular (2 points)
- Intracellular concentration of potassium and proteins is higher than extracellular (2 points)
- Ion channels and sodium-potassium pump located in the membrane (2 points)

The movement of ions is presented correctly (6 points in total)

- Potassium flows outside the cell through the potassium channels (1 point)
- Sodium flows inside the cell through the sodium channels (1 point)
- The sodium-potassium pump pumps 3 sodium ions outside the cell and 2 Potassium ions inside the cell (2 points for correct movement of ions, 2 points for correct numbers of ions)

Scoring SimSketch model (13 points in total)

Divide the score by 1,3 for grading.

The model contains the following structures (1 point for each structure, 3 points in total)

- Ligand gated (clickable) sodium channel
- Voltage gated sodium channel
- Voltage gated potassium channel

The settings of the voltage gated ion channels are correct (4 points in total):

- Sodium channel opens: when the membrane potential is higher (more positive) than a negative value (1 points) at least higher than the membrane potential at the start of the simulation (1 point).
- Potassium channel opens: When the membrane potential is higher (more positive) than a positive value (1 points), at least higher than the value of the sodium channel (1 value).

It is possible to simulate an ‘action potential’ with the model (6 points in total):

1. When the clickable sodium channel opens, membrane potential rises enough to open the voltage-gated sodium channel (2 points).
2. When the voltage gated sodium channel opens, membrane potential rises enough to open the voltage-gated potassium channel (2 points).
3. When the potassium channel opens, the membrane potential gets under the value that opens the voltage-gated sodium channel (2 points).

Scoring reflect on modeling assignment

2 points for every meaningful adaptation (every adaptation that would lead to a more realistic or sophisticated model)

6 points for a meaningful application for the model:

- 2 points for describing a meaningful application
- 4 points max for describing the experiment in detail (1 point for every step of the experiment)

10 points in total

Scoring Insight Question

- 2 points for right answer (channels open easier)
- 2 points per reasoning step
    - Opening chloride channels leads to an influx of chloride
    - Influx of chloride leads to a more negative membrane potential
    - With a more negative membrane potential it is harder to generate an action potential (because more sodium has to flow in to arrive at the threshold)
    - Less action potentials will lead to sleepiness.

10 points in total

Appendix C: mechanistic reasoning

Table B1

Elements of Mechanistic Reasoning

Element	Abbre viation	Score	Identifying reasoning elements + example
Teleological or anthropomorphic statements	TS	-1	Students make a teleological or anthropomorphic statement about the target phenomenon. *'The sodium ion wants to go to the other side of the membrane'*
Asking 'How'-Questions	HQ	1	Students propose a question about how the phenomenon works, they ask about the underlying mechanics. *'How does a nerve cell regulate the flow of sodium ions?'*
Identifying Setup Conditions	SC	2	Students identify and verbalize clearly the conditions that make it possible for the mechanism to run. *'The intercellular concentration of sodium ions in a resting nerve cell is lower than the extracellular concentration (so sodium ions will diffuse down their concentration gradient into the cell)'*
Identifying Entities	IE	3	Students recognize specific entities that enable the target phenomenon or mechanism to run. *'The sodium channel allows sodium ions to diffuse across the membrane'*
Identifying Activities	IA	4	Students identify and verbalize the actions and interactions among entities. *'The ions move randomly and bounce off each other'*
Identifying Properties of Entities	IPE	5	Students identify and verbalize general properties of entities that enable the mechanism to run. *'Sodium ions are positively charged'*
Identifying Organization of Entities	IOE	6	Students identify and verbalize spatial or temporal organization of entities that enable the mechanism to run. *'the extracellular concentration of sodium ions is higher than the intracellular concentration'*
Chaining	C	7	Students chain properties and activities of entities and verbalize causal relationships. Backward chaining is hypothesizing how the target phenomenon came into being, it is looking backward in time.

	'An action potential occurs when sodium ions flow into the cell, so their concentration outside the cell was higher than their concentration inside the cell' Forward chaining is hypothesizing how the mechanism will proceed in time. *'When the sodium channel opens, sodium will flow into the cell, making the membrane potential less negative'*

Coding scheme adapted from Russ et al. (Russ, Scherr, Hammer, & Mikeska, 2008)

Adaptation:

'Describing Target Phenomenon' exchanged with 'Asking How-questions'. Because we work with a phenomenon that students are familiar with and that will be introduced by the teacher, describing the target phenomenon is no indication of mechanistic reasoning. Asking 'How-questions' (from van Mill) is a better option, because it indicates if students ask the right questions that, when answered, lead to mechanistic explanations instead of anthropomorphic or teleological explanations.

Last two items (analogies and animated models) not included because they are not hierarchical.

Appendix D: Analyses of reasoning and modelling actions

In this document the reasoning processes and modelling actions of the students during the action potential assignment are analysed and compared. In the assignment students first have to come up with a mechanism for action potential, so they reason together about this phenomenon. More specifically, the students have to write down what ion currents are happening and which channels are facilitating this currents for three different stages in an action potential: reaching the threshold value, depolarization and repolarization. Next, they have to create a model for an action potential in SimSketch. By analysing their reasoning steps before and during the modelling assignment, it is possible to examine if mechanistic reasoning contributes to modelling performance and to what degree learning has taken place.

Veer and Leon

Reasoning

Leon: It becomes positive so that one goes open. No, that one opens, because then the things will get in. So positive sodium ions get… Depolarization. That is the same, right? That one opens.
Veer: Yes, but it could be possible that both are open here. Here it is more profound than here. So something else should happen. Here it goes… it increases. He goes like this.
Leon: Maybe here the potassium channels open also. So that it only makes a small difference.
Veer: Yes, and first the potassium because they are still open.
Leon: Yes, potassium will also open.
Veer: And then it closes. And that has an effect.
Leon: Yes, open. And here, potassium channel is closed. Sodium channel open. And repolarization is the other way around, then the sodium goes…
Veer: Yes that is the other way around, because then suddenly…
Leon: …Potassium channel open, sodium channel closed.

Worksheet answer

From resting potential to threshold potential:
- Sodium goes in through the opening of sodium channels.
- Potassium channel also goes open.

Depolarization:
- Potassium channel is closed.
- Sodium channel opens.

Repolarization:
- Potassium channel open.
- Sodium channel closed.

Analysis of reasoning

According to Veer and Leon, the initial change in membrane potential (from resting potential to threshold) is caused by the opening of sodium channels. Subsequently, they agree that the potential changes only a little bit compared depolarization. Therefore, potassium channels have to open simultaneously to balance it out. Depolarization occurs when the potassium channels close and the sodium channels are still open. They argue that for depolarization the status of the channels invert, so the sodium channels close and the potassium channels open.

Modelling (and reasoning during modelling)
The students changed the existing 'always open' sodium and potassium channels into channels that could be opened with a mouse click. They tried to model an action potential by opening the sodium channel and the potassium channel by clicking it. Then they were stuck because it was not possible to close the channels by clicking them (they reasoned that first both the sodium and potassium channels were open and for depolarization the potassium channel would close).

Leon: *Oh so it goes open. How can I close it?*

Even after the researcher asked the students for multiple times to think about what triggers opening of the channels and explained that the change in voltage causes the opening of sodium channels, they still used only the 'clickable' function on channels. However, they seem to understand that the difference in rise of membrane potential is facilitated by different channels.

Leon: *Oh so you just put the sodium channels on it. So you have to be able to click on those. Yes indeed. So when the threshold value of minus 50 or something is reached… You have to go to minus 50 for a moment. So if we click on it it goes up when I'm right.*
Veer: *No that is the situation with a stimulus.*
Leon: *Oh so I have to be able to click both of them.*

It is notable that the students talk about a threshold of -50mV that corresponds with the graph, but not with their own model. They created another sodium channel which opens on a mouse click. Because they used double icons (clickable and voltage-gated) on the same object, the researcher interfered with the modelling process.

Researcher: *What do you want to attain with which channel?*
Leon: *I want to make all channels clickable.*
Veer: *Why?*
Leon: *because then you can regulate them and open this one so it goes to the threshold value and then this one so it goes up, and then close all of them and then open this one so it goes down again.*
Researcher: But what is the second step?
Leon: *Oh with those voltage-gated things, but how can we make those?*

Leon now seems to understand at least the first two steps of the action potential, but because he did not know how to make voltage-gated channels he tried to model it with mouse clicks. He also understands that the stimulus is a little flow of sodium ions, and depolarization is facilitated by a greater flow, because he makes the *little* sodium channel clickable and the *bigger* sodium channel voltage-gated:

Leon: *It starts at… minus 8. So when you put it on zero then it will reach the threshold value.*
Researcher: *How do you reach zero?*
Leon: *With the little sodium channel.*

Subsequently, Leon decides on his own that the potassium channel also has to be voltage-gated, so that it opens automatically when the nerve cell is depolarized:

Leon: *No I have to set it, not on clickable but on voltage gated. Because when it for example higher gets then a certain, let's say 30, it goes open.*

After running the model Leon concludes that the membrane potential will never reach +30mV, so he changes the trigger to +15mV. This is the last change made in the model before they ran out of time.

Score of assignments
Hand-drawn model pretest: 3,0
SimSketch model: 6,9
Reflect on modelling assignment Leon: 2,0
Reflect on modelling assignment Veer: 2,0
Insight Question Leon: 2,0
Insight Question Veer: 0,0

Conclusions

- Veer and Leon reason *mechanically* about the phenomenon.
- They construct a hypothetical mechanism *together*.
- They use their new knowledge of ions and ion channels in a nerve membrane *coherently*.
- Their hypothetical mechanism sounds *logical* and could be theoretically correct.
- The mechanism is not explained fully by their hypothesis because they do not mention what triggers opening or closing of the channels.
- They do not mention different types of membrane channels → this is a sign that they do not understand how these channels might function on a *molecular* level. This might be the result of the model itself (*focusing effect*), because the model focusses on ion flows and keeps the molecular functioning of the membrane channels a *black box*.
- Their first model is *coherent* with their reasoning. At first they made both channels clickable and tried to open them both and then close the potassium channel. Only because this did not work (closing on a mouse click is not possible in the software) and the researcher explained them that some channels open on voltage, they created another sodium channel, but still clickable and not voltage-gated.
- Leon seemed to have *learned* something because he changes the original plan and starts to work with multiple voltage-gate channels and one clickable sodium channel. From the discussion it is not clear whether Veer is still following what he is doing.
- In the end the students *successfully* created a model that could visualize the three main steps of an action potential. However, a lot of help from the researcher was necessary for them to proceed.

Stijn and Isabelle

Reasoning
Stijn: *Depolarization arises from positive sodium ions that can enter suddenly. So first the channels are closed, so you only have that sodium potassium pump maintaining the resting potential. And suddenly, by an external stimulus those positive sodium ions can enter so the interior of the cell becomes positive. You can see that the membrane potential rises, it becomes positive.*
Isabelle: But the external stimulus that…? Isn't that a little…? Oh, but where do the positive sodium ions go?
Stijn: *Well, those are just inside on a certain moment and the potassium ions will go outside and the sodium-potassium pump restores how it was before.*
Isabelle: Oh, okay.
Stijn: *And this all happens very quickly because the refractory period, the period in which that is not possible for a moment is very short, so it all happens very quickly. Okay.*

Worksheet answer:
From resting potential to threshold potential:

- Sodium ions go inside
- Sodium channels that are always open

Depolarization:

- Sodium ions go inside

- Sodium channels that open at -50 mV

Repolarization:

- Potassium goes outside
- Potassium channels that open at +30 mV

Analysis of reasoning

Stijn starts the discussion by explaining what happens during depolarization: suddenly positively charged sodium ions flow into the cell and thereby making the inside of the cell more positively charged. According to him, the sodium-potassium pump is the only active membrane protein during resting potential, which is in line with the informal model of this dyad. He seems to ignore the previous experiments that demonstrate that leaking of ions through membrane channels is necessary for maintaining the resting potential. Isabelle points out that he did not explain what causes the opening of sodium channels and indicates that she does not understand where the sodium ions go. Subsequently Stijn mentions that the sodium ions are inside the cell and potassium ions outside, and that the sodium potassium pump then restores the beginning situation. They did not mention different types of channels or what triggers opening or closing of the channels.

Because the students only described the ion currents and not the type of membrane proteins on the worksheet, the researcher asks what type of channels facilitate these ion currents. Still Stijn only answers: ‘sodium channels’. Therefore, the researcher explains the different types of channels and asks the students to think about what triggers opening of the channels. This is very difficult for Stijn and Isabelle to comprehend, only after explaining three times the different channels they started to discuss the matter. At first Stijn ignores Isabelle, but she is the first to introduce that they need another sodium channel in order to model an action potential, and that they have to think about the role of the potassium ions:

Stijn: *These are the channels that are always open, these are….*
Isabelle: *those are triggered by voltage, right? But what about the potassium?*
(Stijn is writing, does not react)
Isabelle: *Okay and next we have to make another sodium channel?*
Stijn: *I think so.*

Eventually they wrote down a mechanism that partially explains the action potential:
From resting potential to threshold is facilitated by sodium channels that are always open.
Depolarization is facilitated by voltage-gated sodium channels that open at -50mV.
Repolarization is facilitated by voltage-gated potassium channels that open at +30mV.

Modelling (and reasoning during modelling)

In line with Isabelle’s last comment, the students draw another sodium channel. This voltage-gated channel opens at -50mV, so they copied this value from the graph. They start a discussion on whether to set it ‘higher than’ or ‘lower than’ 50 mV, because their model’s resting potential is around 0 mV. Isabelle seems to focus on their own model, while Stijn only refers at the provided graph:

Isabelle: *Yes that’s what I said! Because the cell starts at -2 or something, so it is already above -50.*
Stijn: *Yet it has a resting potential of -70, right?*

The researcher mixes in and points out that they have to think of values that fit with their SimSketch model. He also mentions that they can choose themselves if they want to change the membrane potential of their model, by changing the amount of charged particles. Stijn decides to make more negatively charged proteins.

The students have no idea what the next step should be, so the researcher helps them again by explaining that the cell still needs to be stimulated, and asks the students what this stimulus could be. Stijn seems to know exactly how this works in a neuron:

Stijn: *a neighbouring change of membrane potential or a neurotransmitter.*

Next, the researcher explains that they could provide a stimulus with a 'clickable' channel. Again, Stijn shows he understands the relationship between the size of the channel (and thus the flow of particles) and the membrane potential:

Stijn: So it stay closed until you click on it. Okay so you just make another channel.
Isabelle: What kind of channels is this?
Stijn: It also is a sodium channel. Make it a little bit bigger, they are allowed to overlap a little. Yet maybe not to big because those are only little changes, right?

So they drew another sodium channel which could be opened by clicking on it. After running the model it occurred to Stijn that the threshold for the voltage gated sodium channel is not realistic, so they change it to -10 mV. Their membrane is really crowded with ion channels, partly because the screens they were working on had a very low resolution and because they kept all the 'leakage' channels from the previous assignments. Therefore, the researcher suggests to think about what channels they really need for the action potential, whereupon the students make the leakage channels smaller and the necessary channels bigger.

After changing the threshold of the sodium channel to -5 they are able to let this channel open after opening the clickable voltage channel. The students get enthusiastic from seeing this chain reaction:

Stijn: *Come on, reach it! Yes it is open.*
Isabelle: *Yeah!*
Stijn: *But they are streaming out a lot.*

Subsequently the researcher mixes in with their discussion to make them think about repolarization. Stijn and Isabelle answer together:

Researcher: *and how are you going to facilitate the next step?*
Stijn: *That is when the positive potassium...*
Researcher: *Suddenly it seems to drop drastically, right?*
Isabelle: *So in fact then you have to add another potassium channel so when the voltage reaches a certain…*
Stijn (interrupting Isabelle): *and those have to be closed also, right? So those have close at a certain voltage and those have to open at a certain voltage.*

So they did draw a potassium channel and let it go open at +10mV. This was decided by Isabelle:

Isabelle: *Shall we put that one on 10 or something? Because it did reach that value some times already.*

The next adaptations are: making the leakage channels even smaller and set most of the thresholds lower in order to make the action potential more likely to happen. When the researcher announced that it is almost time, Isabelle seems determined to make the mechanism work:

Isabelle: *I think that we put that one too high, shall we make it a bit lower for one more time? I just want it to work!*

Score of assignments
Hand-drawn model pretest: 5,0
SimSketch model: 8,5
Reflect on modeling assignment Stijn: 4,0
Reflect on modeling assignment Isabelle: 4,0
Insight Question Stijn: 10,0
Insight Question Isabelle: 2,0

Conclusions

- At the beginning of this assignment, Stijn proposes a mechanism that does not explain the phenomenon, because he only mentioned the ion currents and did not say anything about specific ion channels. However, he did reason mechanically.
- After the researcher explained the different types of ion channels, they come up with a mechanism that is nearly complete, they only neglected the sodium channels that open at a stimulus.
- The students did not really read the list of hints for a quick start.
- Although Stijn takes the lead during most of the reasoning, Isabelle often questions his statements or comes up with something Stijn did not think of. This results in efficient cooperation.
- The students were stuck in the beginning of the modelling assignment because they had no idea how to start an action potential. This is in line with the gap in their reasoning.
- Stijn and Isabelle got enthusiastic when they were able to simulate the first two steps of an action potential. Subsequently, they really tried to make the model work as good as possible and focused on creating a smooth chain reaction of opening and closing channels.
- Their final model does explain action potential sufficiently.

Arie and Ruben

Reasoning
Arie: *Let's see, one, then it is just evenly distributed. Two.*
Ruben: *No it is closed until the threshold value, or until it is stimulated so intense that all of them will open. Then they open tremendously.*
…
Ruben: *There are three settings: always open, mouse click and trigger less than, greater than. The channel must open when the voltage becomes greater than -50 millivolt. This is action potential, so the threshold value… So it has to open when it… When both of them are closed nothing happens.*
(Students click around in SimSketch, exploring channel settings)
Ruben: *All right, resting potential is -50, but we don't have that, the resting potential is zero, minus two in this case. But that's impossible because…*
Arie: *Resting potential is -70.*
Ruben: *Yes but when there are closed it does not change. They only open when… Okay they have to open when… The thing becomes lower? Higher than… And when that happens the whole action potential goes.*

Worksheet answer
The channels have to go open when the voltage reaches the threshold value.

Analysis of reasoning
Ruben takes the lead in these fragments and at first he gives two brief explanations of the mechanism of action potential: 1. It (the sodium channel) is closed until threshold is reached (and

then they open exceedingly). 2. They (the sodium channels) open when they are sufficiently stimulated. When the students have read the assignment fully and have learned about the three types of ion channels, Ruben start anew with reasoning: When both channels are closed, nothing happens, the channel has to open when the threshold of -50 mV is reached. He seems not so sure if the channels must open when the thing (membrane potential) gets higher or lower than the threshold.

After and during this episode of reasoning, the students check out the membrane functions in SimSketch without discussing a specific goal behind this. Subsequently, the researcher motivates them to think about the mechanism that is responsible for the first two *different* phases in an action potential (from resting potential to threshold, from threshold to depolarization).

Ruben: *Okay, Phase one is closed.*
Arie: *First only sodium goes. Only potassium is closed.*
Ruben: *The stimulus will close the channels, that is a mouse click. And when the cannels open, sodium is green, so than the particle will become more positive. So then, when it gets even more positive then have to open more, or something?*
Arie: *Yes more of them will open.*
Ruben: *So you need two channels or so.*

In this fragment the students cooperate better. Their new explanation is that first only a sodium channel opens and the potassium is still closed. A stimulus opens the channels, the particle gets more positive, and when it is more positive, more channels have to open. Although the students' reason is inaccurate (particles getting more positive?), Ruben notices something important: you need two channels.

It is remarkable that their reasoning seems to be focused on the software and less on the provided visual model from BINAS.

Modelling (and reasoning during modelling)
It was difficult to follow the students' modelling actions exactly because of some problems with the log file and because they changed a lot of settings in a trial and error like way. However, the researcher saved their final model so it is possible to see where they got.

Halfway the previously mentioned reasoning episode, they changed the existing 'always open' sodium channel into a clickable sodium channel. This was done randomly, as is concluded from the following fragment:

Ruben: *I have no idea what this will do.*
Arie: *What did you do? When you click it they will open?*
Ruben: *Yes, but that does not matter.*

After the reasoning episode, they created another sodium channel and set it to open at 7mV, which is in line with their reasoning:

Arie: *We will make a second channel.*
Ruben: *Always, no only when it is greater that a positive value of voltage.*

It took them some time and they ran the model for several times to get all the setting right. It is not completely clear from the log file, but it is plausible that they now have the three different channels that you need in order to create an action potential: a clickable sodium channel, a voltage gated sodium channel, and a voltage gated potassium channel. Next, the students try to fine-tune the model in a logical way:

Arie: *But I think the sodium channel will close right now.*
Ruben: *Okay, oh it should definitely stay open for a longer time.*
Arie: *No but is should clode when the potassium channel comes open*

During the modeling process they notice that they've overseen something: how does hyperpolarization take place?

Arie: *But how does hyperpolarization takes place?*
Ruben: *Because way more particles will go out than go in. Because of the pump.*
Arie: *So the pump it restores after a while, right?*
(students are observing the model)
Ruben: *Why doesn't it make it sink back?*

Interestingly, they hold on to their belief that the sodium-potassium pump is able to make the membrane potential negative. In experiment 4 and 5, they kept hanging onto the idea that this pump makes the membrane potential negative because it pumps two positive ions in the cell and three positive ions into the cell, even though they've done experiments with it and concluded they were wrong. However, Ruben seems to understand that their model is not complete:

Arie: *Hey, what a stimulus!*
Ruben: *Wow! But hyperpolarization doesn't take place.*

Subsequently, the students are absorbed in playing with the zoom-function, so the researcher tries to inspire them to create a more accurate model by pointing out the negative membrane potential and distribution of particles in a real nerve cell. The students choose to change the resting membrane potential by making more negatively charged proteins and by adapting the thresholds of the voltage-gated channels to the new situation.

During the last minutes of this assignment the students show that they know the function of the potassium channel in hyperpolarization. Ruben first shows that he understands that there is a link between the potassium channel and hyperpolarization:

Arie: *So those will endlessly go open, the potassium?*
Ruben: *No, those quit eventually, when it's hyperpolarized again.*

Later, Arie points out that opening of the potassium channel depends (threshold: +24mV) on how many sodium ions are likely to flow into the cell:

Arie: Yes, and what is the charge of one particle?
Ruben: One.
Arie: How many are there? This way it will never reach 25, you understand that?

Their final model contains the necessary ion channels to model an action potential. However, the voltage-gated sodium channel is smaller than the clickable channel, and the threshold of the potassium channel is too high to reach with this model.

Score of assignments
Hand-drawn model pretest: 5,5
SimSketch model: 6,9

Reflect on modelling assignment Arie: 2,0
Reflect on modelling assignment Ruben: 3,0
Insight Question Arie: 6,0
Insight Question Ruben: 10,0

Conclusions

- There are no signs of higher-order mechanistic reasoning in the initial reasoning of Ruben and Arie. Moreover, they use only half sentences and do not really react at each other during their initial discussion. They did not come up with a well-described mechanism.
- The students did not discuss the provided information in detail.
- There was a lot of 'trial and error' behaviour during this modelling task, the students lost a lot of time.
- The students tried to find answers and logic for the target mechanism in the software (by checking drop-down menu's etc.), without really discussing their reasoning beforehand.
- During their second discussion of the mechanism (after the researcher made them reason about it again), they did cooperate better and their discussion led to new insights that were implemented in the model (two different sodium channels for both stages).
- By observing the model, Ruben learned that their model was incomplete and they missed a mechanism to hyperpolarize the membrane. Later in the process, he mentions that hyperpolarization is facilitated by the potassium channel. This is proof that he actually learned something about the target phenomenon from using the modelling software.
- There final model contains the necessary channels, but it does not explain action potential fully.

Ewout and Karel

Reasoning

Ewout: *Opening and closing of the channels, so apparently they can open and close.*
Karel: *Yes, just by closing and opening them. Yes when for example this one closes the potential, then it gets more positive. And when this one closes here it gets more negative.*
Ewout: *And when both close than it does not change.*
Karel: *Yes, well probably it will. Because this one is three and two.*
Ewout: *Oh. So then it will slowly become more negative.*
….
Ewout: *Yes at a certain voltage it has to open, right?*
Karel: *Yes but wait a minute, I think that we just need this one and this one* (student refers to SimSketch)*. Because you don't need this one for action potential, right? These two are the most important. So only sodium and potassium channels.*
Karel: *Indeed, I think that at a certain voltage, at a certain voltage these have to… Because look, at trigger this one suddenly gets very high, so after is this one has to come open. When it's high this one has, and after it becomes too low so then this one has to go open. You know, so it goes like this.*
Ewout: *All right, let's check if that's correct. As a result of the threshold the sodium channels open.*
Karel: *And then this one becomes really high. And then in the inside it becomes really high. And during depolarisation it becomes even higher, so during repolarization it becomes lower.*
Ewout: *Yes so during a certain high voltage these have to come open.*
Karel: *Yes, that is the potassium.*
Ewout: *No, that is during a very low voltage. No sorry at a high voltage.*
Karel: *And at the threshold value sodium channels. I it is correct like this.*
Ewout: *and the pump takes care of this?*
Karel: *Yes.*

Worksheet answer:
K- and Na-channels.
At a very high voltage, K-channels open.
At threshold, Na-channels open.

The mechanism suggested by Ewout and Karel is very close to the textbook explanation: Only the potassium and sodium channels play a role in this process (not the sodium-potassium pump). At threshold, sodium channels open with a high potential (more positive) as a result. During depolarization, it gets even higher. During repolarization, K-channels open because of a high (more positive) voltage. Their explanation lacks a clear difference in the difference between an small change in membrane potential (from resting potential to threshold potential) and the actual depolarisation.

Modelling (and reasoning during modelling)
The students start with very small channels, which they drew in the previous assignments. Karel and Ewout change the functions of the already existing sodium and potassium channels. The sodium channel is changed into a voltage-gated channel which opens at -50mV. Ewout points out that this is way too low for their actual model, so they change it into -5mV. The potassium channel is also made voltage-gated, and opens at +3mV.

After running the model, the students recognize a problem: how does the membrane potential reach the threshold that opens the sodium channel?

Ewout: *It doesn't do anything, no it I stuck at -2.*
Karel: *Yes but it should change soon. And then it should become -5.*
Ewout: *Yes, but how could it change?*

The researcher explains that there are three different types of channels (always open, clickable, voltage-gated). Karel seems to understand that they need another sodium channel in order to facilitate the two different influxes of sodium ions:

Karel: *Okay look we set this trigger on 'greater than' and then just that one. We make another one that opens on a mouse click.*

However, instead of making two different sodium channels, they change the voltage-gated channel into a clickable channel. While running their new model, Ewout simulates multiple stimuli by repeatedly clicking on the sodium channel.

Karel: *Don't click so often!*
Ewout: *No but sometimes there are multiple ones, sometimes one stimulus is not enough.*

Subsequently, they change the open time of the sodium channel. After running again they make the potassium channel smaller because it 'makes it too negative'. They also changed the number of charged particles (40 of each ion, 4 proteins). They run the model again and start a discussion about the resting potential and the role of the potassium channel in restoring the resting potential, without really getting somewhere.

Ewout: *But by this thing it becomes lower I think.*
Karel: *Yes this one only makes it worse.*
Ewout: *That's stupid!*
Karel: *So then maybe we have to make this one a bit bigger? Or smaller, so that he doesn't make it lower, no wait!*

The researcher notices the stagnation in the modelling process, so he asks how they differentiate between a stimulus and what happens after reaching the threshold. This triggers a productive discussion in which Karel reminds that he mentioned earlier they needed different channels, and Ewout to propose features of the channel.

Karel: *That's what I said, right? That you make one that opens on a click and one that opens on that threshold.*
Researcher: *Okay, try it out!*
Ewout: *That one has to be greater than.*
Karel: *Yes this one has to be open only for a short time.*
Ewout: *Yes, but long enough to reach the threshold. This one is already good.*

So they draw another channel and choose to let it open at a voltage of -5mV. The researcher points out that they are working with really small channels which makes exchange of ions very slow. The students rearranged the channels and made them bigger. They ran the model again and decided a lot of settings have to be changed. So they changed the thresholds of both the sodium and the potassium channels. From the discussion it may be concluded that the goal of the adaptations is to recreate an action potential as it is described in the BINAS model:

Ewout: *Yes, look now this one goes open, and this one should already be closed so it was open for a too long time.*
Karel: *Yes it should be lower. This one should be way lower.*

Moreover, the students did become enthusiastic because the model behaves like they want to: in the audio record Ewout says: 'And now we have some sort of a peak, and then it goes like this, and now...' followed by yelling and clapping. Subsequently, they ran the model several times and adapted channel settings quickly.

Score of assignments
Hand-drawn model pretest: 4,5
SimSketch model: 10
Reflect on modeling assignment Ewout: 5,0
Reflect on modeling assignment Karel: 3,0
Insight Question Ewout: 2,0
Insight Question Karel: 8,0

Conclusions

- During their initial discussion of action potential, the students reason together and mechanically. They come near to a text book explanation.
- The gap in the students' explanation of the phenomenon (no difference between stimulus and depolarization) also showed up in their model (only one sodium channel). So their modeling actions represented their initial reasoning.
- By observing the model, the students recognized this gap and learned that they needed different ion channels.
- Ewout and Karel started with very small channels which slowed down every process they wanted to simulated. They changed it only after the researcher suggested bigger channels.
- The students got enthusiastic while watching the first successful chain-reaction of opening and closing channels.
- After all necessary elements were implemented in the model, Ewout and Karel efficiently fine-tuned channel settings.

- It is possible to simulate an action potential with their final model.