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Teaching New Methods

A cognitive and sensory approach based on workshops using the zebrafish model promotes the discovery of life sciences in the classroom

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Abstract

The main objective of the ZebraCool programme was to create a positive attitude and curiosity towards science by bringing experimental activities within schools using an introductory cognitive and sensory approach. This innovative programme was offered at all levels of primary and secondary education including vocational high schools. Thematic workshops can be carried out on various themes such as comparative anatomy and embryology, molecular biology and evolution, or toxicology and endocrine disruptors. They were on an ad hoc basis or as part of an annual school project using zebrafish as a model. This animal was a very attractive entry point for the educator to motivate students to appreciate biology, in particular in the field of molecular biology and evolution. For each practical workshop, the student was an actor in his/her learning, which was intended to arouse the curiosity and desire to understand and learn. The programme was based on close collaboration between class teachers and programme educators to adapt workshops' content to the school curriculum. Students conducted their own experiments, formulated and tested hypotheses, learned laboratory techniques, collected, and analysed data. ZebraCool scientific activities fell within a conceptual framework of evolutionary biology through which participants perceived their own inner fish through the comparison of biological processes between humans and zebrafish.

Keywords: cognitive and sensory approach; biology education; teacher–researcher partnership; zebrafish

Introduction

The state of STEM teaching

Given the lack of scientific literacy among their populations, education systems across the world have become aware of the difficulty many professionals have in teaching science and the lack of enthusiasm among pupils for these subjects [1, 2]. Facing the need for internationally comparable evidence on students' performance, the Organisation for Economic Co-operation and Development (OECD) launched the Programme for International Student Assessment (PISA) in 1997. PISA assesses the extent to which 15-year-old students have acquired key knowledge and skills essential for full participation in social and economic life [3]. Science scores tended to decrease after 2012, but have remained stable since 2018, while reading and mathematics scores have dropped drastically [3]. France figures among the OECD's countries that face a fall in science, technology, engineering, and mathematics (STEM) education. A recent report from the French Academy of Sciences states that there is a lack of skills to teach science subjects in primary schools [4]. Indeed:

> The majority of teachers have degrees in literature, humanities and social sciences, history, geography and languages. Only 14% of primary school teachers have followed a scientific university education, even if more than a third hold a scientific high school diploma. This is a major cause of their difficulties, giving the feeling that the world of science frightens school teachers with a lack of confidence and skills in teaching these subjects. [4].

Studies about teachers teaching science topics within and outside their areas of specialism highlight important differences in the quality of preparation and delivery of science lessons [1, 2, 5–7]. When teaching outside their area of expertise, teachers express anxiety, apprehension, and a lack of confidence in facing the challenges of teaching. Moreover, this lack of teacher confidence can be transmitted to students with the idea that the world of science is inaccessible. This can lead to difficulties in making science interesting and less stressful for students [8, 9]. When the teaching of science begins in elementary school on a weak and very theoretical basis, consolidation in middle school is needed to foster discovery of the different scientific disciplines and provide adolescents the gift of scientific insight to better understand themselves and the world around them. The implementation of the problem-based-learning (PBL) approach in the teaching of science may not only alleviate teachers' apprehension, but also enhance students' motivation and comprehension [10, 11].

Furthermore, there is a significant lack of connection between secondary education and the higher education system. High

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school students have only limited exposure to scientific careers and degrees, and little is done to make these study programmes attractive and accessible. To remedy this and help teachers stimulate students' interest in science, several countries have developed educational programmes involving scientists, leading to a positive impact on pupils' behaviour towards science [12, 13]. In addition, such collaboration between teachers and researchers improves teachers' ability to teach inquiry-based science, as well as their confidence and scientific knowledge [10, 13]. In the USA, the Science Education Partnership Award programme [14], e.g. funds innovative STEM and informal educational projects for school years 1–12 aiming to improve student understanding of health sciences. In Europe, the Partnerships For Science Education [15] programme develops digital learning packages for public health education and PBL application. These projects connect a wide range of actors including schools, universities, non-formal education providers, enterprises, and civil society organizations, and engage them in efforts to enrich STEM education. In France, 'La main à la pâte' and 'Les P'tits débrouillards' are the main national programmes designed to popularize science, make it accessible to the largest number of students, and assist teachers in bringing STEM projects into schools. Even though scientists participate in these associations, they do not provide access to academic research. Through these two programmes, students have few opportunities to discover scientific research and talk to researchers. Given this state of affairs, and as we are convinced that clear communication on research jobs and university programmes could help students to envision a scientific career, we decided to create the ZebraCool programme.

ZebraCool uses zebrafish to make STEM teaching more attractive

Initially partly inspired by the BioEYES project [16], the ZebraCool programme is a non-profit organization created in 2017 by Ms. Laure Bourcier and Pr. Patrick J. Babin at the University of Bordeaux, France. The programme relies on the expertise of the team members on the zebrafish model to intervene in schools. The ZebraCool programme is available for all levels of primary and secondary education as well as for vocational high schools. This programme utilizes an aesthetic and sensory approach (this approach is explained in the subsection below), particularly for young audiences, as a starting point for discovering biology, including molecular biology and evolution. Additional information about the French education system is given in Supplementary data (Supplementary Table S1). In 2020, ZebraCool received the approval of the French Ministry of National Education to intervene during class in support of teaching activities in public schools and to participate in the development of educational research. The association uses zebrafish as an attractive entry point for children and adolescents to open them to the scientific approach and stimulate their interest in science.

The zebrafish model is extensively applied in biological science, but zebrafish are ideal organisms for education, as well [16, 17]. The list of characteristics that make zebrafish an excellent classroom model is long. Zebrafish are cost effective, easy to maintain, and very reliable at producing large numbers of embryos on demand. Both the embryos and early larvae are semi-transparent and can be viewed under a stereoscopic microscope facilitating manipulations and detailed observations during development. Several programmes already employ zebrafish at school to foster STEM education: BioEyes (USA) [16], InSciedOut (USA) [18], or Zebrafish University College of London (UK) [19]. Furthermore, the website Zebrafish in the Classroom [20] is intended to serve as a valuable resource for both teachers and students who are utilizing zebrafish in undergraduate courses. It provides protocols for numerous common techniques, including but not limited to raising zebrafish, producing embryos, conducting classroom experiments with live zebrafish, and conducting data- and image-based virtual experiments.

The content of the ZebraCool workshops described below is adapted to the school curricula according to the classes involved, and adjusted in consultation with the teachers of those classes. The programme aligns with national science education standards, teaches basic scientific principles, fosters critical and creative thinking, requires students to effectively collaborate with peers, and introduces science- and health-related careers. Workshops alternate interactive experiences and classroom discussions. The topics addressed concern, e.g. comparative anatomy, reproduction, development, genetics, evolution, physiology, and toxicology, including endocrine disruptors. Supervised within the classroom by teaching and research professionals, most of the workshops consist in direct observation of and experiments with living zebrafish, the goal being stimulation of curiosity and a critical scientific spirit. This hands-on style of education can be very challenging for teachers involved in our workshops, but it provides students with an opportunity to experience scientific research guided by researchers.

From an educator's perspective, ZebraCool incorporates age-appropriate methods, requires efficient class management, aligns objectives and standards with attainable goals, and uses interdisciplinary approaches to foster cooperative student-centred learning experiences. Studies have reported that participation by scientists in such programmes enhances their communication, mentoring, and teaching skills, while also enabling them to serve as student role models [7, 13].

ZebraCool workshops illustrate that we can use the scientific method of reasoning in our daily life to understand the world we live in, i.e. stating a problem and hypothesis, gathering and analysing data, discussing and formulating arguments, forming and providing evidence for claims, as well as reviewing and providing feedback. PBL is an approach to teaching in which students address real-world challenges, through an inquiry-based instructional method, to accomplish meaningful projects, thus engaging in knowledge construction. Inquiry-based science education (IBSE) has been proposed as a framework for creating a learning environment in which we observe a shift from teacher to student-centred class design and development of peer-teaching, peer-assessment and PBL [11]. It is claimed that IBSE has the potential to provide students with authentic experiences of how scientists work [21]. At ZebraCool, our aim is for students to gain experience with a model species and practice a wide range of experiments. This work is presented in scientific and technical, but popularized language, in a more interactive format than traditional textbooks and theoretical classes. The implementation of the workshops is intended to increase the qualification and confidence of the teachers to approach scientific topics. The investigational workshops mobilize interdisciplinary learning and transversal skills among students, who may develop their ways of thinking, reasoning and acting by cultivating oral and written language [11, 22]. Retrospective studies on the students' academic development, even at the elementary school level, have demonstrated that students' results and intrinsic motivation to learn science improve through hands-on experiments. Moreover, the interaction with external research contributors has been reported to benefit teachers in terms of enhancing their educational skills [11, 16, 23–26].

Conceptual framework and emotional usefulness of the zebrafish

Aesthetics may be defined as the science of sensitivity, which studies the nature and perception of beauty [27]. One of the main aims of aesthetics is to study the experiences and judgments that arise when we perceive the world. This enables us to classify something as attractive, ugly, sublime, elegant, etc and to express emotions. The emotional status can be influenced by sensory information such as visual information, taste, and touch. Educational cognitivism can make use of sensory elements as learning tools in an effort to activate student motivation, understanding and memory.

Many authors have described aesthetic aspects involved in teaching science [28]. Aesthetics, in general, has two interrelated meanings: (i) a set of design and art practices, (ii) the responses of affect, emotion and taste to experiences and objects, regardless of the practice, which also includes scientific endeavours [28]. Seeing a purpose and a meaning can generate aesthetic-related feelings, which many hold to play a substantial role in meaning-making and learning [29, 30]. Scientists have repeatedly pointed out that, similar to art, science has its own type of aesthetics [31], which can be revealed through experimentation, e.g. This concept is based on Dewey's work in which he advocated placing experience and practical activities at the heart of teaching methods [32, 33]. Connecting experimental lessons to everyday-life concepts and objects helps students to conceptualize science and feel closer to it [34]. The aesthetic approach to teaching science partially relies upon interconnecting fields in order to stimulate students' senses and emotions regarding sciences. Indeed, Wickman [29] and Lemke [30] have hypothesized the importance of students' feelings and perceptions in the meaning-making process and the role of the latter in learning effectiveness. Studies supporting these hypotheses have demonstrated the preference of students for experimental science classes, and the effectiveness of emotions induced by demonstration and laboratory work on student's attention and learning [35–38].

The start of any ZebraCool intervention with students is to have an aesthetic and poetic vision of what a zebrafish represents (Figure 1). It is a very beautiful living being from which pupils can learn about the world of fish, but also about themselves by observing what distinguishes us from them and what resembles us, i.e. our own inner fish [39]. The general principle underlying ZebraCool workshops based on the biological model is to make it clear that what we observe today is the result of a long evolutionary history of life on Earth. What both unites us and separates us from zebrafish is a phylogenetic link from which we can comprehend the evolutionary conservations and subsequent innovations made from the speciation event that separates us from our common ancestor. This general vision is introduced during the implementation of the workshops and is declined with more or less details depending on the educational level of the participants. The soft first approach is to observe the zebrafish in its aquarium, the beauty of its shape and skin colour, the elegance of its swimming and movement in groups, and the anatomical difference between the females and males. From the observation of its external appearance, we then invite the student to discover its anatomy in a more precise way. Using a stereomicroscope, the interior of its body is observed at different stages of the development of its embryo and larva. The semi-transparency of these early stages of development inevitably leads children to the enchantment of seeing, e.g. the heart beating and the blood circulating. This type of intellectual stimulation is at work whatever the age-group, adults included. This emotional approach to designing our relationship with this animal partner will facilitate scientific curiosity and involvement in the implementation of the workshops.

In your fish-tanks, in your ponds,
Zebrafish, how you live for aeons!
Does death forget you,
Fish of melancholy.

Figure 1. Sweet dreams and beautiful reality (adapted and translated from a poem by Guillaume Apollinaire, The Bestiary: or Orpheus's Procession, 1911). The serenity of a child drawing a zebrafish.

Implementation of the ZebraCool programme

Material, animals, and human resources

ZebraCool provided dedicated equipment, e.g. quality binocular magnifiers including cameras, as well as small laboratory equipment and reagents for carrying out experiments. ZebraCool also furnished teaching supplies on how to draft protocols, analyse results, and interpret them. Along with adult zebrafish, ZebraCool provided batches of eggs, embryos, and eleutheroembryos. Zebrafish rearing being straightforward, teachers who wished to do so had the possibility of installing an aquarium in their class prior to an intervention. For this, ZebraCool supplied fish rearing equipment, animals and a zebrafish husbandry guide. Upstream school teacher training was conducted for implementing the workshops as well as learning to rear zebrafish, when applicable (Supplementary Fig. S1). Teacher training included a very precise description of the rearing conditions to be respected in terms of photoperiod, water temperature, food and density of animals in stalls. In addition to the zebrafish rearing guide, laminated summary sheets were provided for posting next to the aquariums.

All the procedures used for animal breeding and reproduction were conducted in compliance with the European Communities Council Directive (2010/63/EU) and the local French legislation (Ministère de l'Agriculture et de l'Alimentation) on the protection and care of animals used for scientific purposes under permit number A33-522-6. Wild-type zebrafish were made available by the MRGM laboratory, University of Bordeaux, France. ZebraCool only used live zebrafish that were less than 5 days post-fertilization in age for observation under the stereomicroscope. Because of their very early stage of development, these eleutheroembryos were not subject to animal experimentation regulations. The adults used came from the reclassification of animals from the MRGM laboratory. These animals were only used for breeding in class, when this option was chosen, and for making observation drawings of adults. The animals used to observe the different stages of development were previously fixed in 4% paraformaldehyde and supplied by the laboratory. The biological samples from adult animals, i.e. the scales, were taken from salmon fish skin provided by a local supermarket as it is considered food waste. Under no circumstances were students authorized to carry out actual animal experiments. Information on animal experimentation, the animal welfare regulations that govern it, and the 3Rs rule (refinement, reduction, and replacement) was provided during the workshops in an age-appropriate manner.

ZebraCool provided human resources to transfer their scientific and educational expertise. Educators were professors and assistant-professors, researchers, and technical staff from the local University and public research institutes, post-docs, but also PhDs or master's degree students. The interventions were under the supervision of the ZebraCool scientific council in terms of educational content and methods that were used.

Intervention methods used in the class

ZebraCool's programme was offered at the different levels of primary and secondary education, as well as in vocational high schools. Whatever the type of school, the interventions were available in the form of a workshop or a combination of several workshops. For elementary schools, an annual school project was possible. In that case, all the players in the school's educational community were involved and all classes in the school were enrolled in the programme with several periodic interventions per class during the same school year. Before each intervention, a close collaboration was established with elementary school teachers or middle- or high-school biology teachers. ZebraCool strived to instore a long-term partnership with teachers in order to allow the sustainability of interventions over several years within the same school. Each type of workshop, defined by its theme, was offered with content and complexity adapted to the age groups of the children or adolescents involved and by integrating it into the programme of each school cycle.

A ZebraCool intervention could be scheduled on an ad hoc basis for a minimum of 2 hours, or repeatedly within the same class during the school year in the case of an annual school project. The workshops were designed for students to build the knowledge necessary to describe and understand the world around them and develop their reasoning capacities. ZebraCool proposed twelve types of workshops in terms of thematic content (see details in the section below). Workshops with the same theme used increasingly elaborate, abstract, and complex ideas depending on the educational level of the participants. With the help of the educators, the students were immersed in a scientific investigational process: questioning, observation, actual or virtual experimentation, description, reasoning, and conclusion.

An intervention in an elementary school took the form of rotating thematic workshops with five to six pupils per group. For each workshop, documents were given to students to complete with the collected information. These documents were created in collaboration with the teachers of the class according to the level and skills of the pupils. For example, students in early grades of elementary school were often asked to apply stickers with little writing, while older children typically wrote or drew observations with precise annotations.

For secondary schools, interventions within a class were often in the form of a multi-thematic module, i.e. the association of several thematic workshops, with a duration of about two to three hours. Workshops were designed as investigations to solve, alternating theoretical and practical activities. Groups of two or three students were formed. They were given time periods to explore and collect clues to answer the questions, and time to debate about the clues and validate the answers all together. It was important that all the students understood the responses to each phase of the investigation, so that they would be able to pursue the game. With this in mind, we strove to ensure an educational progression of the whole class. At the end of each intervention, a feedback session was organized. Participants could ask any questions that were not answered during the workshops. All biological topics could be discussed in connection, more or less, with the workshops. We believe that this free speaking time was very relevant from an educational point of view and it appeared to be greatly appreciated by the students.

An annual school project was a formula adopted by several elementary schools and typically involved ZebraCool interventions for all classes in the school. The students had to manage the animal husbandry in the classroom over several months under the supervision of the teachers. This responsibility meant that each student took turns feeding the animals and cleaning the aquariums, which tasks were carried out successively by groups of students formed within the class. This also included long-term observation of the fish behaviour. In a school, it was easy to set up a time frame to allow for a longer-term project with, e.g. an intervention twice a year for classes from Years 2–6 (UK educational system). This was one advantage of observing living organisms: it allowed observations and conclusions to be monitored according to the development of the organism studied. Between the workshops, ZebraCool educators and the school's teachers worked together to create a common thread around the zebrafish throughout the year, including connected class projects led by teachers, such as plastic arts activities, geography, ecology, and so on. Figure 2 illustrates two plastic arts activities developed during a school year project: Fig. 2A shows a decorative fish fresco of an elementary classroom around the theme 'Today I am … '. Students drew a fish and labelled their drawing to indicate how they felt that day. This type of activity enabled students to express their emotions while learning how to represent a teleost fish, i. e. what were its attributes. Through such activities, it was hoped that children might also discover that drawing can be a non-verbal channel of communication to express themselves. Figure 2B represents a Year 6 classroom decorated with three dimensional fish constructed by the students. Projects like this were intended to instil students with pleasure and pride as they entered the classroom that they themselves had decorated, thus making the environment more pleasant and stimulating to learn in. Plastic arts activities associated with fish husbandry were expected to enhance the accessibility and enjoyment of science lessons by illustrating these stunning living animals that could be directly observed within the classroom (Fig. 2C). This gentle and progressive approach to the scientific process was designed to enhance thinking, memorization, learning, and use of language.

Thematic scientific workshops and their aesthetic and sensory approach

As noted above, ZebraCool offered twelve thematic workshops. Here we describe the twelve scientific concepts and how they involved aesthetics, the senses and emotions to teach biology and how the lessons included interdisciplinary projects in some cases.

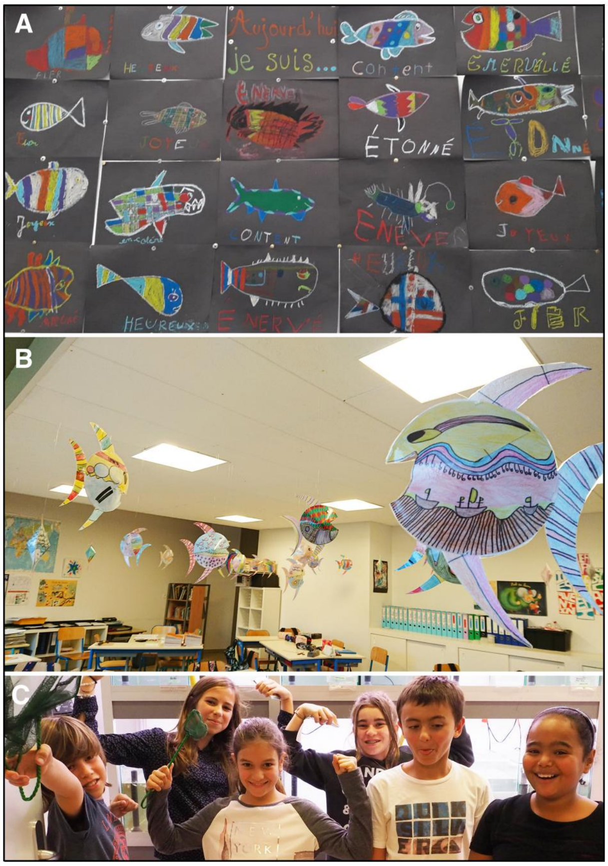

Figure 2. ZebraCool is fun and lets us express our inner fish. (A) Decorative fish fresco around the theme 'Today I am … '. Students drew a fish and labelled their drawing to indicate how they felt that day: e.g., proud (*fier*), happy (*heureux*), pleased (*content*), amazed (*émerveillé*), angry (*en colère*), surprised (*étonné*). (B) An elementary school classroom decorated by the children to illustrate the fish theme. (C) A group of students motivated to learn more about zebrafish.

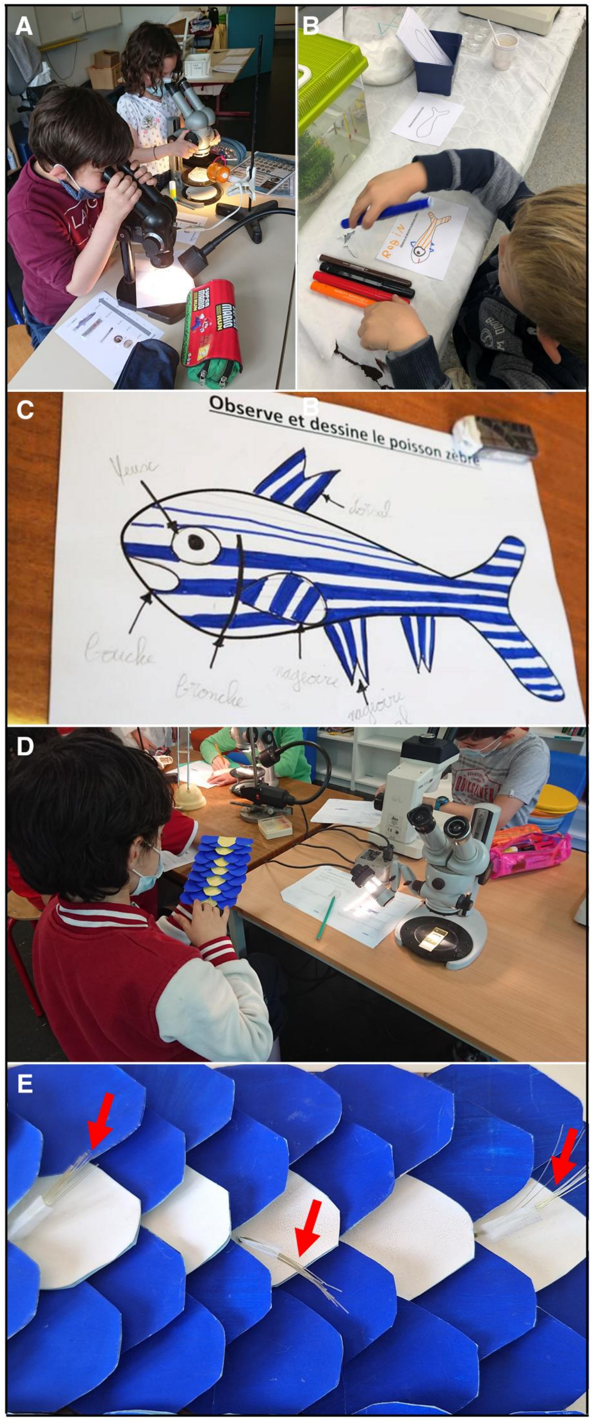

Figure 3. The pleasure of discovering the anatomy and development of zebrafish during elementary school workshops. (A) Recognize and classify different developmental stages of zebrafish using observation under the stereomicroscope. (B) Draw an adult zebrafish from an animal observed in its aquarium and learn to distinguish males from females. (C) Annotated drawing of an adult zebrafish. (D) Observing the scales of zebrafish. The comparison of what is observed under the stereomicroscope with a model made of paper that the child holds in his or her hands. (E) Cardboard model of part of the skin of the adult zebrafish with its covering scales and the sensory hair cells of the lateral line neuromasts (red arrow).

Thematic workshops description

Workshop n°1: Comparative anatomy and embryology (Year 2-13/CP-3$^{\text{ème}}$). The first goal was to describe the organization of the body plan and the presence or absence of certain organs and tissues in zebrafish compared to humans. The second goal was to describe the successive stages of development of these two species, and to place this description in an evolutionary perspective. Students might thereby learn about their own body and organs: Where are they located? What do they look like? What are their functions? As a striking and concrete aspect of anatomy, a fanciful plastic skeleton was used to illustrate the human body. The same colour code was used on zebrafish diagrams to illustrate common organs and structures addressing the concept of species evolution.

Workshop n°2: Sexual reproduction (Year 2-13/CP-3$^{\text{ème}}$). The objective was to provide elements of description and understanding of the reproduction of organisms using zebrafish as a starting point. Depending on the class level, discussions were initiated on the different reproductive strategies that exist in animals, including the human species. In front of an aquarium with adult zebrafish, students were asked to make an observation drawing of the animals (Fig. 3B, C). Zebrafish were used to explain the concept of sexual dimorphism based upon their beautiful colours: females present blue and white stripes with a rounded abdomen while males have blue and orange stripes and a more elongated body. These visual characteristics allowed to directly identify the animal's gender. Then, we described how zebrafish mate to explain reproduction of this species. The practical application of this concept was illustrated by direct observation of zebrafish embryos under a stereomicroscope.

Workshop n°3: Development, growth, and nutrition (Year 2-13/CP-3ème). The main objective was to describe the functions related to development and growth that lead to the formation of an adult organism, but also to discover the functions of nutrition of a living being. The zebrafish was used as a model from which comparative explanations could be given for other animals, including the human species. The practical activity consisted in observing the different stages of development of zebrafish, from early embryonic to larval stages, which last several days (Fig. 3A). By observing living embryos, it was possible to see them moving in their 'envelope' called the chorion, and also to observe the movement of red blood cells, e.g. Observing a semi-transparent larva under a stereomicroscope almost inevitably appeared to elicit from children, whatever their age, a sense of wonder that kindled their curiosity and desire for understanding. They also discovered the migration of embryonic cells and the elaboration of anatomical structures with a kinetic video of zebrafish development. Depending on the class level, the pupils were invited to create a flip booklet representing the development of the zebrafish. Creating such booklets mobilized artistic and craft skills that were intended to help pupils to remember what they learned and gave them a tool to share their knowledge.

Workshop n°4: Zebrafish skin (Year 4-6/CE2-CM2). The objective was to understand the organization and role of fish skin as a protective and sensory organ (Fig. 3D, E). This activity started with an observation drawing of a fish scale using a stereomicroscope to understand how scales are composed. Pupils were then introduced to a model of fish skin allowing them to run their hand back and forth over the surface to feel how the scales overlap each other to protect the zebrafish body. The cardboard representation of the adult zebrafish skin consisted of covering scales and the sensory cilia that protrude from the surface along the lateral lines (Fig. 3E). Children could feel how their hand displaced the cilia, fostering better understanding of the role that these cells play in the fish's perception of mechanical waves, sounds, and water currents. The children's own sense of touch on the surface of the cardboard model was then discussed and compared with the analogous sensory perception in fish. The perception of sound and mechanical waves by sensory hair cells in the ear and neuromasts was also discussed. Moreover, the colours used on the model helped to address the subject of pigmented cells and their functions. Made from paper and cardboard, this model could also serve as a class art project that included a scientific modelling aspect following a construction protocol.

Workshop n°5: The heart and blood circulation (Year 2-13/CP-3ème). This workshop covered the composition, functioning and role of the cardiovascular system. A video support from a French science outreach programme showed the heart mechanism and how blood cells circulate through the vascular system in living zebrafish [40]. Students learned to assess their pulse by touching their forearm or throat. The change in heart rate during physical exercise performed by the participants was then compared with that of the zebrafish larvae. The physiological rationale for this modification then seemed more ripe for discussion. This exercise was intended to promote pupils' connection with their own bodies and feelings to better understand what goes on during physical activities.

Workshop n°6: The principle of dilution with coloured chemistry (Year 2-6/CP-CM2). This was an introduction to the basics of laboratory chemistry: dilutions of coloured solutions and manipulation of laboratory equipment for sampling and measurement. Calculating dilutions also involved simple mathematics. Dilutions were prepared using methylene blue to give a highly visual effect (Fig. 4A). Children were also introduced to a sensory approach to the principle of dilution by combining colour and taste. They conducted a serial dilution of strawberry syrup in water before drinking it in order to correlate the decrease in colour intensity with the reduced intensity of the strawberry taste (Fig. 4B).

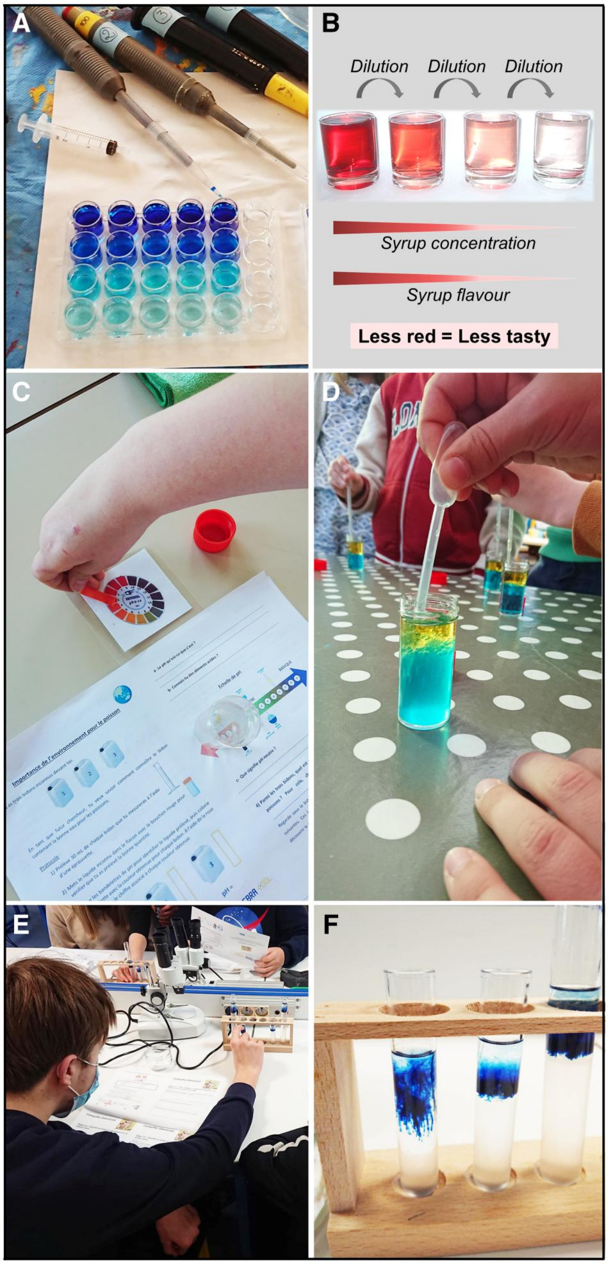

Figure 4. It is nice to experiment, but what do the colours means? (A) The principle of dilution with coloured chemistry workshop. Methylene blue serial dilutions in multi-well plates. (B) Dilutions of strawberry syrup in water to correlate the decrease in colour observed with the decrease in taste according to the dilution made. (C) The pH of water workshop. Using pH paper to measure water pH used for fish rearing. (D) Impact of pollution on the food chain workshop. Experiment that mimics the impact of an oil spill on aquatic organisms through the modification of air/water gas exchanges due to the hydrophobic nature of oil. (E, F) DNA extraction performed during the genetics workshop.

Workshop n°7: The pH of water (Year 4-6/CE2-CM2). This practical workshop consisted in collecting water samples to measure pH and making comparisons with the pH of a zebrafish aquarium (Fig. 4C). In the case of an annual school programme, the presence of an aquarium within the class enabled teachers to work on themes such as water quality by taking regular readings of physicochemical parameters. Students learned about the concepts of acid-base and pH, how water quality affects aquatic organisms, and how to measure volumes and collect samples following a protocol. More than fish biology, this was an interdisciplinary workshop involving Math, Ecology, Physics and Chemistry. The use of colours on the pH scale coupled with measurement of daily-life products, such as vinegar and soap, was designed to promote understanding of the pH principle.

Workshop n°8: Impact of pollution on the food chain (Year 4-6/CE2-CM2). This workshop studied the impact of human activity on the environment and living species. This phenomenon was explained using a scale model with various stickers to mimic an ecosystem and demonstrate the bioaccumulation of environmental pollutants and their transmission through the food chain, from algae to the fish consumed by humans. The use of a simple and colourful scale model allowed the concepts to be integrated as the activity progressed. The workshop also included a quick virtual experiment simulating the effects of an oil spill on ecosystems to understand its potential impact on aquatic organisms and human health (Fig. 4D). Using colour differences among various compounds, this workshop simulated the impact of an oil spill on aquatic organisms through the modification of air/water gas exchanges due to the hydrophobic nature of oil.

Workshop n°9: Genetics (Year 10-13/3$^{\text{ème}}$-terminale). This workshop illustrated the basis of genetic information and the molecular evolution of species. Conceived as an investigation, the goal was to identify an unknown species after extraction of its DNA, using a specific-coloured dye to reveal and observe it (Fig. 4E, F). (For more detail on the scenario, see the combined workshops section below).

Workshop n°10: Cladistics and phylogeny (Year 10-13/3$^{\text{ème}}$-terminale). The purpose was to illustrate the unity and diversity of biological organisms, the use of different criteria to classify species and identify relationships between organisms, and provided conceptual bases for understanding their evolution. The starting point for this workshop was a set of documents given to students for them to complete a table of characteristics shared between zebrafish and humans. Information about zebrafish had to be collected by the observation of several developmental stages under a stereomicroscope.

Workshop n°11: Toxicology and endocrine disruptors (Year 10-13 and vocational high schools/3ème-terminale, lycées professionnels). This workshop was intended to foster an awareness of the impact of endocrine disruptors, including obesogens and neurotoxicants, on human and environmental health (Supplementary Fig. S2) [41, 42]. Through documentary research and games, participants were taught to recognize toxicants found in our environment, as in food, cosmetics, household products, as well as contaminated water and air (Supplementary Fig. S2A–D). The effect on health was illustrated through research carried out using zebrafish (Supplementary Fig. S2E) [43–46]. By examining common cosmetic products, this workshop established a connection with everyday life, to elicit the students' emotions and reactions, in an attempt to render the subject more captivating and intriguing (For more detail on the scenario, see the Supplementary data).

Workshop n°12: Scientific research professions (Year 2-13/CP-terminale). The main aim of this workshop was to show that research is an accessible environment where everyone has a place, including from a professional point of view. A virtual tour of our laboratory was offered in a short video to familiarize viewers with a research laboratory that includes zebrafish rearing facilities. Through games and testimonials from researchers, technicians, engineers, and PhD students, participants were introduced to the daily life of a research laboratory and the diversity of professions linked to scientific research.

Combination of workshops

Combined workshops explored specific, more complex themes than thematic workshops alone and lasted from two to three hours. Directly linked to school curricula, they aimed to illustrate the concepts addressed and apply them to concrete examples. The following list is not exhaustive. N°1: Sexual reproduction and development (workshops n°2 and 3); N°2: Comparative anatomy, development and evolution (workshops n°1, 3, 5, and 10); N°3: Genetics, development and evolution (GDE) (workshops n°1, 3, 9, and 10) (Fig. 5); N°4: Simple chemistry experiments (workshops n°6 and 7); N°5: Environmental toxicology and human health (workshops n°8 and 11). These combinations were designed as investigations to solve in order for students to adopt a position as scientific researchers. In the next section, we describe an example of a combination workshop, while highlighting the aesthetic approach used to address scientific knowledge and methods.

Detailed combined workshop N°3 entitled 'genetics, development and evolution (GDE)'

This workshop combined the thematic workshops n°1, 3, 9, and 10. Certain steps of the investigation were implemented in a practical way by the participants, and others carried out in a virtual manner. Educational documents and the notebook completed by students during one such study are available in the Supplementary data section (Supplementary file documents). The scenario unfolded in the following steps:

1. Investigative context and initial observations: The researcher was hosted by local citizens during a vacation in Indonesia, on the island of Java (Fig. 5.1). There was no running water in the house. In the bathroom, there was a drinking water tank with fish in it that seemed to all belong to the same species, as well as small fish larvae (Fig. 5.2). Faced with this curious discovery, the researcher posed a series of questions: Why are there fish in the water supply? What is this species and how can it be distinguished from other species? Are there similarities and differences between this animal and us? How did it get there? Do the animals in the water tank need to be fed and, if so, what do they eat?
2. Species identification by morpho-anatomical study: The researcher did not have a cell phone, camera or internet access, so he or she decided to make the most precise drawing possible of one of the fish for species identification via a bank of fish images upon returning home.
3. Species identification by molecular biology: In parallel, the researcher delicately cut off a fragment of the caudal fin of one of the fish (Fig. 5.3–5.4). This did not kill the fish and fin cuts regenerate in just a few weeks. This sample was not taken from zebrafish during the workshop. To get enough material, we used fish flesh bought at the supermarket. Once back home, the researcher extracted the DNA from

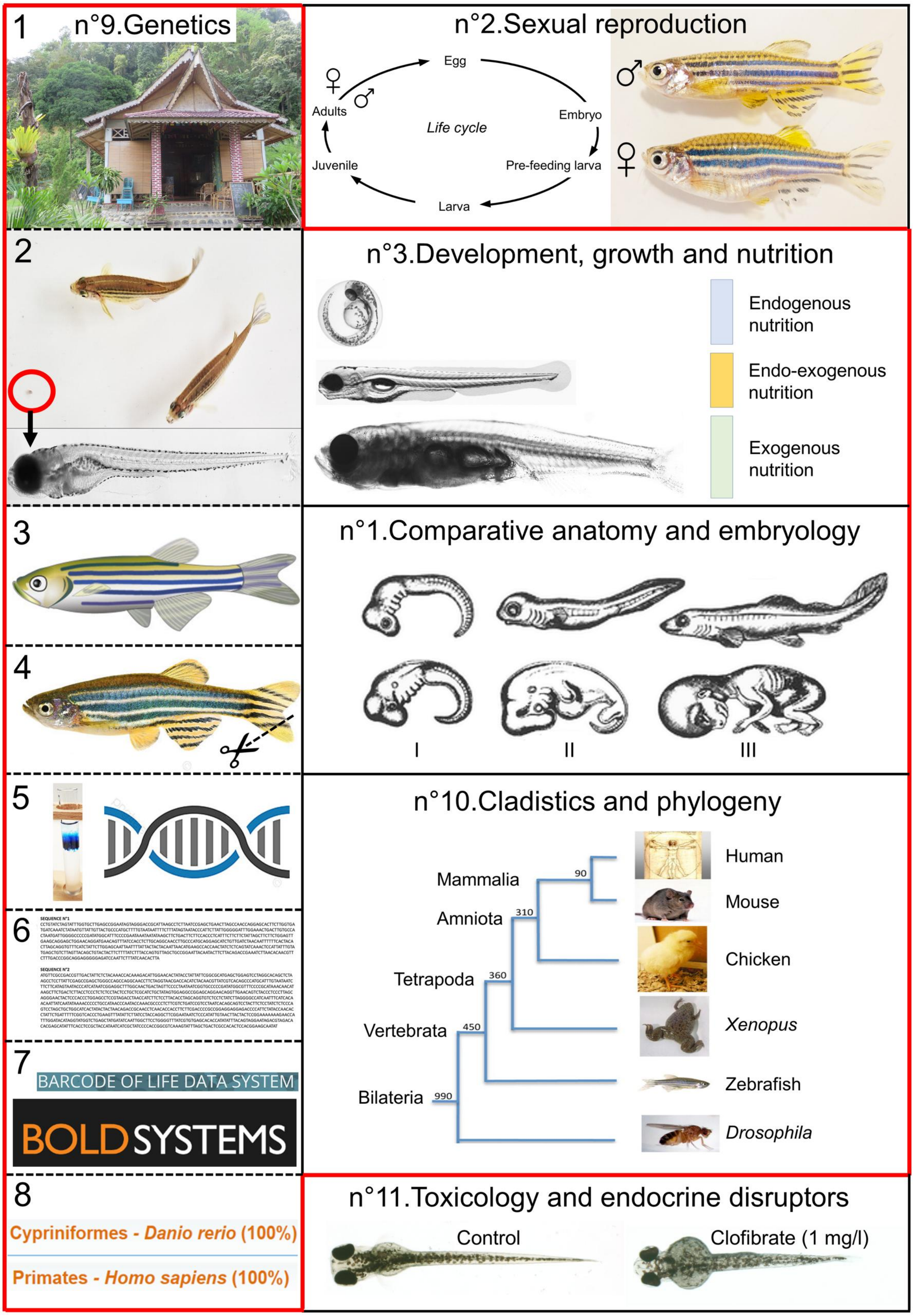

Figure 5. Modular architecture of the ZebraCool programme. Table illustrates the design of the combined GDE workshop. *Left panel*: Dotted black lines define the steps of the Genetic workshop (n°9). *Right panel*: Full black lines partially surround two of the twelve thematic workshops using the zebrafish model; red lines surround the four thematic workshops that constitute the combined GDE workshop (n°1, 3, 9, and 10). See also Supplementary file documents: Educational presentation used by the ZebraCool educators and Students' notebook for the GDE combined workshop. Additional workshops can be associated, such as sexual reproduction and/or toxicology and endocrine disruptors to propose a broader multi-thematic activity, if desired.

the biological sample. A practical illustration was carried out in class with the extraction of DNA from fish flesh and from human saliva taken from the participants of the workshop (Fig. 4D, E). Then, the researcher contacted a molecular biology laboratory and had the DNA barcoding method implemented on the biological sample 'taken in Indonesia'. This method was based on the recognition of a DNA fragment of a given target gene (mitochondrial cytochrome c oxidase I region (mtDNA COI)). The DNA was amplified by the polymerase chain reaction (PCR) method to get a large number of copies. Each living organism has its own sequence of this gene, allowing its identification. The researcher connected to the BOLD identification system server [47] (https://www.boldsystems.org/), a dedicated database, to identify the species to which these two DNA barcodes belong (Fig. 5, left panels 6–7).

4. Data analysis: The result obtained indicated that two species were identified from the biological sample taken on site. The first was a teleost fish, the 'zebrafish (*Danio rerio*)', the second was the human species (*Homo sapiens*). How could we explain the DNA barcoding result showing the human species? (Answer: the 'sample was contaminated with human DNA during collection').
5. Learning about zebrafish: To pursue the investigation, the researcher wanted to collect information about this identified fish species. For that, students observed the developmental stages of zebrafish under a stereomicroscope to study its anatomy (cf thematic workshop n°3) and learned about its development (Fig. 5, right panel).
6. Cladistic analysis and phylogenetic tree: Students were asked to study a batch of illustrated boards that reflect the unity and diversity of biological organisms in order to complete a table of shared characters between zebrafish and humans (cf thematic workshop n°10). A discussion was then opened on the evolution of species and the concept of a common ancestor. The investigation ended with construction of a phylogenetic tree of the studied species (Fig. 5, right panel).

During this combined workshop, students alternated between analysis, experimentations, and whole-class feedbacks in order to pursue their research. Experiments proposed were based on visual effects to foster students' interest and facilitate the comprehension of what they were studying (DNA dye, stereomicroscopic observations, living fish). The use of factual samples like fish flesh and pupils' saliva to extract DNA and visualize it was intended to help make DNA a real concept. Indeed, students often appeared amazed by the possibility of observing their own DNA. Moreover, DNA extraction was all the more effective if pupils had already carried it out in class on parts of other living organisms like onions or bananas. In addition to furthering the discussion of the role of DNA as the source code of living organisms, DNA extraction also served to link the previously studied biology lesson to the virtual investigation during the workshop. Moreover, the study of evolution and phylogenetic links between organisms consistently appeared to elicit surprise and curiosity among students. Indeed, before the workshop students did not believe that we are so close to others animals in terms of shared characteristics and common genes, e.g. The comparison between early stages of embryonic developmental stages between zebrafish and humans illustrated how close we are from an evolutionary perspective. This was a real discovery for students, provoking numerous questions.

Discussion

The classroom ZebraCool programme generated interest and discussion by both students and teachers, within the school as a whole and at home. As an example, results of a post-annual project assessment using a survey questionnaire are provided in Fig. 6 and Supplementary data. Students responded that their classroom had been converted into a research laboratory and that they subsequently considered experimental biology as fun to learn through this hands-on method. Carrying out experiments seemed accessible to them (Fig. 6B) and scientific reasoning could be implemented. As an example, the direct observation of a beating heart and the circulation of blood in the vessels of semi-transparent zebrafish embryos and larvae consistently appeared to fascinate and stimulate the attention of all age groups. Also, pupil reactions to zebrafish rearing in class were overwhelmingly positive (Fig. 6B).

ZebraCool was intended to stimulate pupils' interest in science, but also to help teachers gain confidence in STEM teaching by providing accessible and easy-to-perform educational activities primarily linked to real-life issues (Fig. 6A). Studies have shown the important role of teachers' attitudes in giving pupils a taste for science [48, 49]. A PBL teaching strategy combined with an explicit aesthetic dimension might generate new approaches to teaching and learning science, by making scientific concepts fun, concrete, and applicable. In addition, linking STEM courses to other disciplines such as art, ecology, sport, etc in order to open up and illustrate the courses in a reciprocal manner may help to foster meaning-making that enhances learning [50].

When designing the workshops, we were careful to give concrete meaning to the scientific concepts covered. We believe that effective STEM learning can be enhanced by stimulating the sensory organs (sight, taste, touch, hearing) associated with modelling biological processes (body models, chemical reactions, biological and chemical interactions in the environment) and self-projection in the proposed activities (human skeleton, human DNA, sport, everyday cosmetics). During workshops, students manipulated, experimented, made mistakes, asked questions, and observed using their own senses, emotions, and sensitivity. By taking an active part in the lessons, pupils experienced first-hand what they were learning, likely making the concepts covered more meaningful. As our educational documents were simple, colourful, contain pictures and games, and were completed throughout the duration of the workshops, students tended to remember them. We are convinced that practical and concrete activities reinforced by cognitive aesthetic aspects, substantially enhanced theoretical biology lessons. What is more, we found that when students already had a basic knowledge of the concepts covered, they could relate the present activities to what they already knew. This appeared to help them to gain confidence in their scientific knowledge, as well as to learn and remember new concepts. Furthermore, involving teachers in the design of workshops permitted us to adapt them to the class level and school curricula, so as to propose more appropriate activities to foster pupils' curiosity about science.

We have not carried out a formal assessment of learning outcomes to measure and compare knowledge gain between students who have or have not had access to these workshops. Such an assessment would be worthwhile to evaluate the short- and long-term impact of the programme. A number of schools became long-term partners in the programme by hosting ZebraCool workshops from one year to the next. This was achieved through a close partnership with elementary school

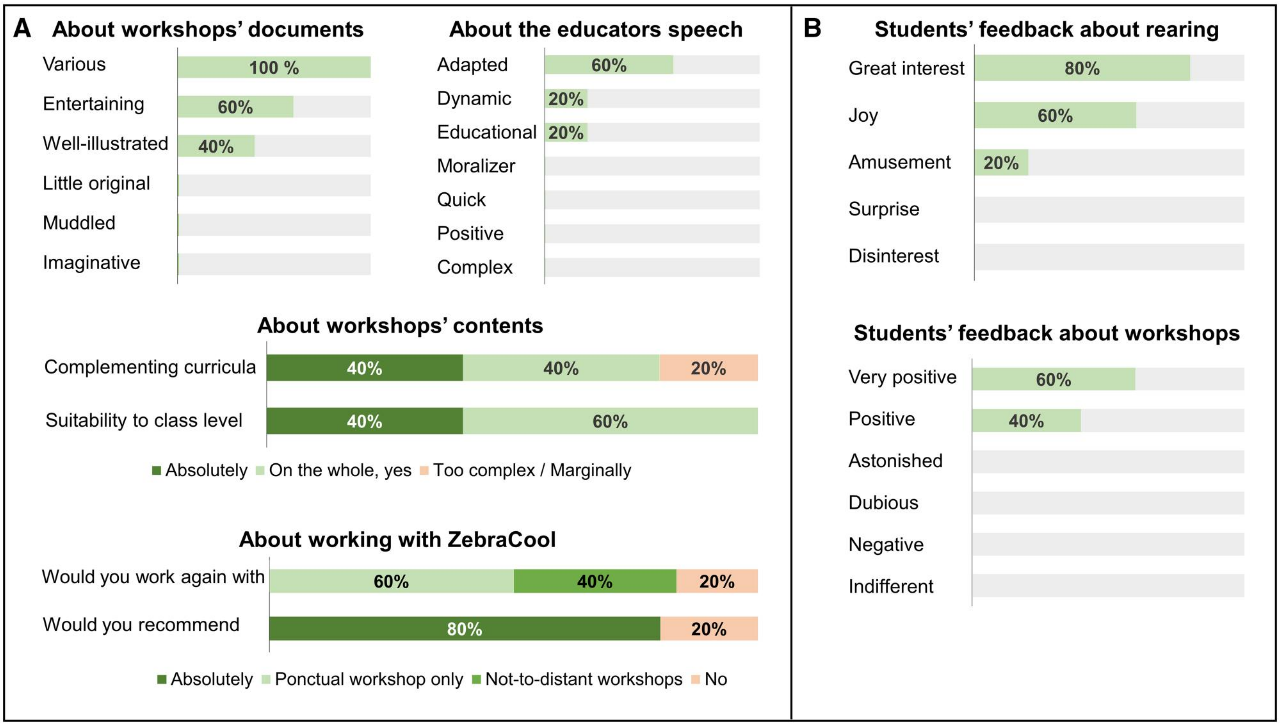

Figure 6. What feelings do ZebraCool workshops elicit? A satisfaction questionnaire was submitted to participants after the workshops (see Supplementary document: ZebraCool survey questionnaire). Example of a post-annual project assessment (see Supplementary document: André Peynaud Elementary School for additional data): (A) Teachers' opinions about the fun and educational quality of the workshops, and whether or not implementing the ZebraCool programme was straightforward. (B) The emotional impact on pupils of zebrafish rearing and the workshops. Eight classes involved, from CP (Year 2/1st grade) to CM2 (Year 6/5th grade), with roughly 25 pupils per class and several presentations per class during the school year. The overall satisfaction rating from teachers following the implementation of this annual project was 4.5/5.

teachers and biology teachers in secondary schools. It appeared relevant and highly motivating for pupils following the ZebraCool program to involve University students (e.g., Master's degree or PhDs) who had spent at least part of their schooling where the workshop took place. In this respect, the testimonies of these speakers during workshop n°12 (scientific research professions) were very inspiring for the high-school students in terms of accessibility to science and scientific careers.

Conclusion

We propose an aesthetic/cognitive approach to initiate or reinforce biology lessons. The aesthetic approach is based upon experiencing first-hand the beauty of nature and science through interactive workshops to stimulate students' interest in biology and foster both learning and memory. The programme is intended to develop a precise sense of observation, promote critical thinking, encourage initiative and teamwork, and may elicit vocations in the fields of science and technology. Finally, more than the knowledge acquired during the workshops, the most important goal is to stimulate curiosity and the pleasure of implementing finely crafted scientific reasoning to discover the beauty of the biological world around us.

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Author contributions

Laure Bourcier (Conceptualization [equal], Investigation [equal], Methodology [equal], Project administration [equal], Resources [equal], Writing—original draft [equal], Writing—review & editing [equal]). Patrick J. Babin (Conceptualization [equal], Funding acquisition [lead], Investigation [equal], Methodology [equal], Project administration [equal], Resources [equal], Writing—original draft [equal], Writing—review & editing [equal])

Supplementary data

Supplementary data are available at *Biology Methods & Protocols* online.

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