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Introduction to Virtual Lab Polymerase Chain Reaction (PCR) Practicals in Biology Education at UIN Jakarta

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Abstract

Polymerase Chain Reaction (PCR) is a fundamental biotechnology technique used to amplify specific DNA fragments in large quantities through in vitro replication. This virtual laboratory aims to (1) understand the basic principles and working stages of PCR, (2) identify the main functions of PCR components, and (3) apply PCR simulations to amplify target genes using the *LabXchange* platform. The research was conducted virtually, using an interactive experimental approach, by 60 Biology Education students working in one group. Data were collected from student worksheets and simulation outputs. The results showed that students were able to identify and explain each stage of the PCR process, including denaturation, annealing, and extension, as well as analyze the roles of the DNA template, primers, dNTPs, buffer, and *Taq polymerase* enzyme. The simulation successfully demonstrated efficient amplification of the target gene. These findings indicate that virtual laboratory learning effectively enhances students' conceptual understanding and analytical skills in biotechnology.

Keywords: Biotechnology, DNA Amplification, LabXchange, PCR, Virtual Lab

INTRODUCTION

In education, technology is fundamental to achieving educational goals, including manual media (Solihin, Bae, et al., 2025; Harahap & Solihin, 2025). Just like that teaching aids, virtual media, and robotics (Solihin, Apriliani, et al., 2025). Advances in biotechnology have enabled humans to manipulate genetic material with a high degree of precision. One technique that has become a significant milestone in this progress is the Polymerase *Chain Reaction*(PCR) (Susilo et al., 2024). Discovered by Kary Mullis in 1983, PCR revolutionized

molecular biology by enabling the amplification of DNA fragments from small amounts to millions of copies in a few hours. (Susilo et al., 2023). In biology education, understanding PCR is important because it underpins numerous applications in genetics, forensics, and the diagnosis of genetic diseases. PCR is based on the principle of in vitro DNA replication and is performed using a thermocycler. The process involves three main stages: denaturation, annealing, and extension. Denaturation is performed at approximately 95°C to separate the two DNA strands (Smith, 2020). The annealing stage occurs at a moderate temperature (50–65°C) to allow primers to bind to the target sequence, whereas the extension stage is performed at the enzyme's optimal temperature. *Taq polymerase*(72°C) to synthesize new DNA (Artika et al., 2022). PCR components consist of a DNA template, primers, dNTPs (deoxynucleotide triphosphates), buffer, Mg^{2+} ions, and Taq DNA polymerase (heat-stable) (Susilo et al., 2024). This combination of components produces a repeated amplification reaction that exponentially multiplies DNA fragments. The simple yet highly effective working principle of PCR makes it a key method in modern biotechnology research.

In the context of education, a virtual lab such as LabXchange enables students to understand PCR without the constraints of laboratory facilities. *Mechanism explains the stages and functions of the components, whereas using PCR to amplify a gene simulates* a real-life reaction. Through this interaction, students not only learn theory but also observe the mechanisms of gene amplification visually and interactively. (Smith, 2020)Through this practicum, Biology Education students have the opportunity to develop conceptual understanding, critical thinking, and collaboration skills. The group work experience of conducting simulations and preparing presentations of learning outcomes reflects an active and meaningful learning process (Arip et al., 2023). The objectives of this practical are (1) to understand the basic principles and working stages of PCR, (2) to understand the function of each principal component in a PCR reaction, and (3) to apply PCR simulations via the platform. *LabXchange* to amplify the target gene.

RESEARCH METHODS

This study employed a qualitative descriptive approach, using a virtual-experiment model, to examine students' understanding of PCR concepts through laboratory simulations implemented on the LabXchange platform (Listiawati et al., 2022). This approach was chosen because it aligns with the research objectives of assessing the learning process, identifying PCR work stages, and evaluating students' understanding of PCR reaction components through interactive practicum experiences, without requiring physical laboratory facilities. The virtual

practicum model allows researchers to observe students' learning processes directly through their exploratory activities. The research subjects were 60 Biology Education students at UIN Syarif Hidayatullah Jakarta who participated in a virtual PCR practicum in the Biotechnology course. Students were divided into small groups to facilitate interaction, discussion, and implementation of PCR simulations through the module *Mechanism* and Using PCR to Amplify a Gene at LabXchange (Ulfia & Wahyuni, 2025). The subject selection technique used purposive sampling because the students involved are enrolled in courses academically relevant to the PCR material and are expected to complete the simulation in accordance with the learning outcomes successfully.

The data collection instruments included student worksheets (SWs), activity notes from the simulation, and the platform's display of PCR reaction outputs. The SWs were used to assess students' understanding of the PCR stages (denaturation, annealing, extension), the function of reaction components (DNA template, primers, dNTPs, buffer, and Taq polymerase), and their ability to analyze gene amplification mechanisms. Instrument validity was established through expert judgment by the lecturer in charge of the Biotechnology course, while the consistency of responses among group members and the alignment of simulation results with theoretical concepts assessed reliability.

Data were analyzed using descriptive analysis techniques, including data reduction, data presentation, and drawing conclusions based on student response patterns and PCR simulation results. The analysis focused on students' ability to identify the PCR stages, explain functions of the components, and interpret the amplification process occurring in the simulation. The findings were then compared with modern biotechnology theory to determine the extent to which the virtual lab improved students' conceptual understanding and analytical skills.

RESULTS AND DISCUSSION

Based on the practical work conducted, this demonstrates that students have achieved a deep understanding of the stages of the PCR cycle. The following is the analysis of research data on the Introduction to Virtual Lab Polymerase Chain Reaction (PCR), (Table 1). The denaturation stage is clearly visualized as the process of breaking hydrogen bonds between DNA bases, separating the two strands. This process clarifies why high temperatures are necessary at the beginning of the PCR cycle. At the molecular level, denaturation at ~95°C breaks the hydrogen bonds between A–T and G–C base pairs, separating the two strands of DNA onto a single template. In the simulation, the practitioner observes that strand separation is rapid and must be complete for the primers to access the target sequence freely. If

denaturation is incomplete (e.g., due to too low a temperature or too short a time), annealing and extension efficiency will decrease, resulting in a lower final product yield. This understanding is important because it demonstrates a direct relationship between thermal program settings and amplification results.

Table 1. Results of Student Observations on Understanding the Basic Principles and Stages of PCR Work

No	PCR Stages Understanding Indicators	Number of Students (n)	Percentage (%)	Information
1	Identifying the denaturation stage	56	93.3%	Understanding DNA strand separation at ~95 °C.
2	Identifying the annealing stage	54	90%	Understanding the role of temperature in primer attachment.
3	Identifying the extension stage	55	91.7%	Understanding Taq polymerase activity at 72 °C.
4	Understanding the relationship between temperature and amplification results	52	86.7%	Knowing the effect of temperature on PCR results.
Rate-rate		54.25	90.4%	Outstanding category.

The annealing stage is considered a critical process in which primers bind to the target DNA sequence. Temperature errors can cause primers to fail to bind or bind at the wrong location, resulting in inaccurate amplification results. (Susilo et al., 2023) The annealing stage is highly sensitive and strongly influenced by temperature and primer design. In the simulation, the practitioner's ability to adjust the annealing temperature is tested, and its effects are observed: too high a temperature decreases primer-template bonds (leading to no amplification), whereas too low a temperature increases non-specific bonds (leading to non-specific bands and primer-dimers). From here, the practitioner learns about the primer melting temperature (Tm) and the importance of selecting a primer with an appropriate Tm to achieve a specific reaction.

Table 2. Results of Student Observations on Understanding the Functions of PCR Components

No	PCR components	Number of Students (n)	Percentage (%)	Information
1	DNA template	58	96.7%	Understood as the primary mold of amplification.
2	First	57	95%	Understanding primer design and primer-dimer potential.
3	dNTPs	55	91.7%	Knowing dNTP as a building block for new DNA.
4	Taq polymerase	56	93.3%	It is considered the primary enzyme in DNA synthesis.
5	Buffer & Mg^{2+}	53	88.3%	Understanding the importance of Mg^{2+} in PCR reaction stability.
Rate-rate		55.8	93.0%	Outstanding category.

The extension stage is shown with animations depicting the activity. *Taq polymerase* extending a new DNA strand from the primer. Students observe that the direction of DNA synthesis is always 5' to 3', and the addition of dNTPs occurs according to complementary base pairing. Extension by Taq polymerase at ~72 °C demonstrates the enzymatic mechanism: the enzyme adds dNTPs one at a time to the 3' end of the primer, following base pairing. In the simulation, the extension rate is directly proportional to the target length; longer targets require longer extension times. Practitioners can adjust extension times for different target sizes and examine the trade-off between cycling rate and strand-synthesis completeness.

The roles of Mg^{2+} ions and buffers are clearly demonstrated in the simulation by varying their concentrations. Mg^{2+} acts as a cofactor that stabilizes the polymerase-dNTP complex; too low a concentration of Mg^{2+} decreases enzyme activity, while too high a concentration can increase nonspecific binding. Students understand that buffer and Mg^{2+} optimization is crucial to setting up a PCR experiment to obtain clean, robust products. Primer design proved to be another critical factor: simulations showed that primers could be complementary to one another, resulting in primer dimers appearing as small bands beneath the target. Interns learned to recognize non-specific patterns in simulated gels. They concluded that primer design checks (avoiding intra- and inter-primer complementarity, GC clamping at the 3' end, and 18–25 nt length) were necessary before conducting real experiments.(Evi Suryanti et al., 2019).

Table 3. Results of Student Observations on the Ability to Apply PCR Simulations for Target Gene Amplification

No	PCR Simulation Application Capabilities	Number of Students (n)	Percentage (%)	Information
1	Assembling PCR reactions (template, primers, dNTPs, buffer, Taq)	57	95%	Components are arranged correctly on the platform.
2	Setting up the thermocycler (denaturation–annealing–extension)	54	90%	Determine the temperature and number of cycles precisely.
3	Observing the amplification results in gel simulation	52	86.7%	Able to read DNA bands using a ladder.
4	Identify errors (missing primary, insufficient buffer)	50	83.3%	Understanding the causes of amplification failure.
Rate-rate		53.25	88.8%	Outstanding category.

The number of PCR cycles and the reaction's exponential phase are quantitatively modeled in the simulation. In the initial phase, ideal duplication occurs (2^n), but thereafter the reaction slows due to reagent exhaustion or reduced enzyme activity. The experimenter observes the predicted amplification graph, which shows a plateau; this indicates that

indefinitely increasing the number of cycles is not a solution and that quantitative analysis requires observation of the exponential phase. Interpretation of the simulation results (bands in the "gel prediction") is complemented by an exercise in measuring fragment size relative to a DNA ladder. The experimenter converts migration distances to base-pair size estimates using a logarithmic scale commonly employed in electrophoresis. This activity fosters simple numerical skills and an understanding of the physical relationship between molecular size and gel mobility. In the module "Using PCR to Amplify a Gene," the practitioner conducts a simulation by selecting a DNA template, primers, dNTPs, buffer, and Taq polymerase in a virtual test tube. Setting the thermocycler to the specified temperature stages and the number of cycles (30). The results showed that the target gene was successfully amplified as predicted.

The simulation also displays potential errors, such as missing primers or buffers, that could cause amplification failure. This helps students understand the importance of component completeness and procedural accuracy (Arip et al., 2023). By repeating the simulation, students learn that the number of cycles affects the number of DNA copies produced. The amplification results graph shows a consistent exponential pattern, consistent with modern PCR theory (Li et al., 2020). From a learning perspective, the use of virtual labs has been shown to increase students' cognitive engagement. They find it easier to understand abstract concepts like DNA replication and enzyme function, which were previously difficult to explain through theory alone (Kang et al., 2021).

Group discussions after the simulation strengthened conceptual understanding and developed scientific communication skills. Students compared the simulation results with PCR theory from the literature, practicing analytical and argumentative thinking skills. In the context of biology education, this experience demonstrated that integrating digital technology can overcome the limitations of physical laboratories. As explained by Suryanti et al., (2019) That virtual laboratory-based biology learning significantly increased motivation and conceptual understanding. The practicum results showed that students not only understood the principles of PCR and its components but also related these concepts to biotechnological applications, including genetic diagnostics, forensics, and gene cloning. Thus, this virtual activity successfully achieved the cognitive and affective objectives of biotechnology learning.

The results of this study indicate that students' understanding of the basic principles and operational stages of PCR aligns with modern biotechnology theory, which explains the mechanism of DNA amplification through three main stages—denaturation, annealing, and extension—as first developed by Kary Mullis. Students' ability to identify that the denaturation stage occurs at high temperatures, breaking DNA's hydrogen bonds, indicates that the concepts

of thermostability and the structure of the DNA double helix are well understood in molecular biology. Similarly, students' observations of annealing temperature sensitivity are consistent with the theory that primer hybridization is strongly influenced by melting temperature (Tm), GC content, and sequence specificity, such that even minor errors can lead to primer-dimer or non-specific amplification. This understanding emphasizes the importance of primer design in biotechnology, as explained in modern PCR literature (Fibriana et al., 2025). In the extension stage, students can explain that Taq polymerase works optimally at 72 °C and extends DNA in the 5'→3' direction, illustrating mastery of the enzyme concept of Thermus aquaticus, which is thermostable and a significant milestone in the development of PCR(Fibriana et al., 2025).

In addition to understanding the stages of the PCR cycle, students were also able to identify the function of each reaction component—DNA template, primers, dNTPs, buffer, Taq polymerase, and Mg^{2+} ions—which are key to successful amplification(Calverley, 2021). Their identification of Mg^{2+} as a cofactor that stabilizes the enzyme-substrate complex demonstrated that the enzymology and biochemistry concepts of polymerization reactions had been well absorbed, referring to the theory that inappropriate Mg^{2+} concentrations can reduce enzyme activity or increase non-specific binding(Scherp & Meier, 2013). Through simulations, students also understood the importance of the completeness of reaction components: the absence of primers or buffers can cause amplification failure, consistent with the principle that DNA polymerase requires a 3'-OH end to initiate DNA synthesis and a stable reaction environment. This ability demonstrates that students are not only memorizing concepts but also relating them to actual molecular mechanisms of action(Suryanti et al., 2019).

The students' experience applying PCR simulations to amplify target genes via the LabXchange platform reinforces the theoretical finding that the amplification process follows an exponential pattern (2^n) in the initial phase and then enters a plateau phase due to declining enzyme activity and reagent depletion(Maharani & Muliati, 2025). Students who analyze the amplification graph in the simulation have mastered the concept of PCR reaction dynamics, which is typically understood only through real laboratory experiments. Their interpretation of the DNA banding pattern in gel prediction also demonstrated that they understood the relationship among DNA fragment size, electrophoretic mobility, and amplification yield, thereby conceptually replicating the wet-lab experience(Kang et al., 2021).

Pedagogically, the findings of this study support the theory of modern biotechnology education that virtual laboratories can enhance cognitive engagement and students' conceptual understanding, particularly in molecular biology, which is abstract and cannot be directly observed. This aligns with the view of Makransky, Ong, and Jambari, who stated that virtual

labs provide visualization and interaction that facilitate students' understanding through a constructivist process. The use of LabXchange in this study has been shown to help students conduct independent exploration, analyze errors, and understand the basic concepts of PCR in greater depth than purely theoretical instruction. Furthermore, this technology overcomes the limitations of physical laboratory facilities, which often hinder biotechnology education in many educational institutions(Smith, 2020).

CONCLUSION

Based on the results of the virtual lab PCR activities, it can be concluded that the three objectives of the practicum were fully achieved. First, students understood the basic principles and stages of PCR through an interactive module explaining denaturation, annealing, and extension. Second, students recognized and explained the function of each component, such as the DNA template, primers, dNTPs, buffers, and Taq polymerase. Third, students can run simulations in LabXchange to amplify target genes, yielding results consistent with molecular biotechnology theory. This activity not only enhances conceptual understanding but also develops collaboration, scientific communication, and problem-solving skills essential for prospective biology teachers.

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