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Abstract. Scientific thinking constitutes a vital component of scientific competencies, crucial for citizens to adapt to the evolving societal landscape. To cultivate students' scientific thinking, teachers should possess an adequate professional knowledge foundation, which encompasses pedagogical content knowledge (PCK). Assessing teachers' PCK of scientific thinking facilitates the development of effective curricula tailored to their continuous professional development. Despite its significance, empirical studies on biology teachers' PCK of scientific thinking are notably lacking. Hence, this research aimed to create a reliable and valid tool to evaluate upper-secondary school biology teachers' PCK of scientific thinking. The results showed that the instrument exhibits high reliability and good validity, affirming its efficiency for investigative purposes. A collective of 292 in-service biology teachers from upper-secondary schools participated in this investigation through the completion of an online survey. The results indicated that, overall, as well as for each component, upper-secondary school biology teachers' performance on PCK of scientific thinking fell within the lower to middle range. Specifically, the performance levels of four components: knowledge of students (KSU), knowledge of instructional strategies (KIS), knowledge of curriculum (KC), and knowledge of assessment (KA) declined sequentially.

Keywords: assessment instrument, pedagogical content knowledge, scientific thinking, upper-secondary school biology teacher

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DEVELOPMENT AND APPLICATION OF AN INSTRUMENT FOR ASSESSING UPPER-SECONDARY SCHOOL BIOLOGY TEACHERS' PEDAGOGICAL CONTENT KNOWLEDGE OF SCIENTIFIC THINKING

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Introduction

Over the past three decades, enhancing scientific literacy has emerged as a primary focus in the field of science education (Han-Tosunoglu & Lederman, 2021). Encouraging scientific thinking is essential for science literacy development in students throughout their education. Learners proficient in science ought to possess the ability to reason in a scientific way and utilize this knowledge in their own and social environments (Krell et al., 2022). *The Upper-Secondary School Biology Curriculum Standards* state that scientific thinking is a crucial dimension of the key competencies (MEPRC, 2020). Fostering the advancement of scientific thinking assists students in gaining a broad range of knowledge and abilities, enhancing their understanding of scientific principles and ideas, and honing their problem-solving skills for practical scenarios (Liu, 2018).

Expanding students' access to various scientific activities and promoting diverse modes of scientific thinking can augment their proficiency in science (NRC, 2010). Numerous national curriculum standards stress the significance of scientific thinking (ACARA, 2018; DfE, 2015; MEPRC, 2020; MoES, 2023; NGSS Lead States, 2013), indicating that fostering scientific thinking is a central goal of worldwide education. Consequently, aligning teachers' preparation with this goal becomes paramount. This underscores the necessity for educators to have a solid understanding of pedagogical content knowledge (PCK), encompassing essential foundational knowledge of the subject matter for effective educational delivery to students of diverse ages, which requires proficiency in teaching techniques, comprehension of complex learning objectives, and insights into students' science learning processes (NRC, 2010). Research on students' learning results and PCK shows a favorable association (Kanter & Konstantopoulos, 2010; Roth et al., 2011; Van Driel et al., 1998), which indicates that the PCK of teachers is essential in nurturing students' development of scientific thinking. However, lower-level cognitive demands still predominate in scientific instruction in schools, even



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though expectations for educational success are shifting to include higher-order thinking abilities like analysis, synthesis, and assessment. (Osborne, 2013). In such scenarios, instructors often maintain control over the learning process, and scientific thinking is not taken into account (Asmoro et al., 2018). Researchers further suggest that teachers need additional training in PCK (Akinoglu & Dilek, 2015; Chen, 2022; McNeill et al., 2016). Shulman (1986) initially introduced and refined PCK. It represents a unique amalgamation of content and pedagogical knowledge, symbolizing a separate type of professional comprehension. PCK denotes teachers' adept understanding and selection of appropriate instructional strategies, modes of representation, and assessment methods tailored to facilitate student learning of specific topics within particular teaching contexts, informed by the learning environment and corresponding curriculum knowledge (Magnusson et al., 1999; Park & Oliver, 2008; Shulman, 1986).

In line with the taxonomies proposed by Veal and Makinster (1999), the PCK of scientific thinking is deemed to be topic-specific. This specificity arises from its focus on particular concepts, terms, or topics within the field, which can vary in instructional styles, approaches, and the depiction of topics under domain-shared concepts. In line with a common recommendation to shift PCK towards concreteness and specificity regarding topics (Loughran et al., 2004; Park & Chen, 2012; Van Driel et al., 1998), the development of PCK instrument was centered on scientific thinking within the upper-secondary school biology curriculum. Biology teachers are tasked not only with understanding scientific thinking but also with knowing how to effectively impart it to students of various ages and backgrounds. While numerous studies have delved into students' scientific thinking, literature provides scant insights into PCK of scientific thinking, revealing a spectrum of varied and individualized viewpoints among teachers. The insufficient rationale behind these metaphors suggests a need for further training in scientific thinking among teachers. Chen (2022) developed and validated an assessment tool composing scale and multiple-choice questions for middle school physics teachers. The results revealed a complex structure of PCK among these teachers, with an overall good level of understanding of scientific thinking. However, teachers in rural areas exhibited relatively weaker performance in this regard.

Research Problem

Beyond fostering conceptual knowledge about the natural world, science education should cultivate scientific thinking like reasoning, explanation, modeling, and argumentation. A variety of research has indicated the essential role of PCK in teachers' lesson design and implementation, learning of new teaching methods, teaching quality, and students' achievement (Baumert et al. 2010; Kanter & Konstantopoulos, 2010; Kulgemeyer & Riese, 2018; Roth et al.2011; Van Driel et al. 1998). As an important dimension of teacher professional knowledge (Gess-Newsome, 2015; Shulman, 1987), PCK ought to be considered a vital indicator of teachers' proficiency and effectiveness (Park et al., 2018). Several scholars have created tools to measure teachers' PCK concerning biological conceptions (Großschedl et al., 2019; Jüttner et al., 2013; Park et al., 2018; Schmelzing et al., 2013). With the advancement of research, the research scope of PCK has expanded beyond the core ideas, such as the nature of science (Faikhamta, 2012) and socioscientific issues (Han-Tosunoğlu & Lederman, 2021). However, few instruments relevant to PCK of scientific thinking are available. Therefore, this study attempted to create a reliable and valid assessment tool to evaluate upper-secondary school biology teachers' PCK regarding scientific thinking. The evaluation and understanding of teachers' PCK of scientific thinking can provide valuable insights for developing effective professional development programs.

Research Focus

From the perspective of science education, scientific thinking is characterized as an intentional pursuit of knowledge, encompassing the capacity to produce, test, and assess hypotheses, data, and theories while also reflecting on the process itself (Koerber et al., 2015; Kuhn, 2010; Zimmerman, 2007). At its core, the fundamental skill of scientific thinking lies in coordinating theories and evidence (Kuhn, 1989, 2002; Kuhn et al., 2008; Kuhn & Pearsall, 2000). Some researchers further delineate the components of scientific thinking, including comparison and classification (Kuhn & Pearsall, 2000), reasoning and explanation (Dunbar & Klahr, 2012; Klahr et al., 2019; Murtonen & Salmento, 2019), model and modeling (Coll et al., 2005; Harrison & Treagust, 2000; Metin & Leblebicioğlu, 2015), argumentation (Asmoro et al., 2021; Kuhn, 1993, 2010), and critical thinking (Azar, 2010; Murtonen & Salmento, 2019).



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Expanding upon Shulman's foundational work, the conception and model of PCK have been refined, translated, and expanded by different scholars (e.g., Geddis et al., 1993; Magnusson et al., 1999; Marks, 1990; Park & Oliver, 2008). Since there isn't a single, accepted definition of PCK, creating a measuring instrument needs to stem from a collective conceptual comprehension of the idea within the field. This ensures the creation of an instrument that effectively guides coherent advancements in PCK research (Park et al., 2018). Pertaining to this, the model presented by Magnusson et al. (1999) was initially adopted due to its widespread use among researchers (Chan & Hume, 2019). However, "orientation" incorporated in this PCK model does not pertain to topic-specific knowledge but rather represents a comprehensive outlook on science instruction, which has garnered minimal consideration as knowledge structures (Nezvalová, 2011).

Consequently, this study focused on four key aspects of PCK: (1) knowledge of curriculum (KC); (2) knowledge of instructional strategies (KIS); (3) knowledge of students' understanding (KSU); and (4) knowledge of assessment (KA). In the phase of instrument design, item contexts will be formulated according to the components of scientific thinking in science education, such as comparison and classification, reasoning and explanation, model and modeling, argumentation, and critical thinking.

Research Aim and Research Questions

This study primarily sought to create and validate a tool for evaluating PCK of scientific thinking among uppersecondary school biology teachers. The research specifically aimed to answer the following questions:

- 1. What evidence substantiates the validity and reliability of measures of the instrument created in this research for evaluating PCK of scientific thinking among upper-secondary school biology teachers?
- 2. What is the proficiency level observed in the PCK of scientific thinking among the surveyed uppersecondary school biology teachers?

Research Methodology

General Background

PCK serves as a critical determinant of teachers' instructional proficiency and is intricately linked with students' academic achievements. Given the prevailing deficiency in fostering scientific thinking within classroom settings, it becomes imperative to measure the extent of teachers' PCK of scientific thinking, encompassing discernible competencies and areas for improvement. This endeavor not only facilitates teachers in engaging in reflective practice but also aids researchers in devising tailored professional development interventions. However, research on PCK of scientific thinking among upper-secondary school biology teachers is limited, necessitating further investigation and scholarly attention. Thus, the present study was initiated to create and confirm the validity of an instrument for assessing this aspect among upper-secondary school biology teachers. This study employed a quantitative approach. After the instrument development, the reliability and validity evidence was collected by expert review, interview and Rash modeling used in a pilot test. Then the revised instrument was used for a formal test. The gathering of data took place between July and April of 2023.

Participants

Convenience sampling was employed to gather diverse participants for this study. Initially, the pilot test involved a selected group comprising 47 upper-secondary school biology teachers, both in-service and pre-service. Following this, a larger cohort of 115 in-service upper-secondary school biology teachers was chosen for the subsequent refinement phase of the instrument. For the formal test, 308 questionnaires were circulated, with 16 being classified as unfinished and thus deemed invalid. Hence, a sum of 292 complete questionnaires was successfully gathered, equating to a 94.8% rate of recovery. Table 1 delineates the basic demographic details of research participants involved in the formal test.



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Table 1

Fundamental Demographic Details Regarding the Participants

	Ν	%
1. Gender		
Male	80	27.4
Female	212	72.6
2. Teaching experience		
Below five years	57	19.5
Five to ten years	37	12.7
Eleven to twenty years	88	30.1
Twenty-one to thirty years	45	15.4
Thirty-one years and above	65	22.3
3. Professional title		
None	28	9.6
Primary	62	21.2
Middle	123	42.1
Deputy senior and above	79	27.1

Table 1, presented above, illustrates the diversity among participants in terms of teaching experience and professional titles. Teaching experience provides insight into the duration of teachers' engagement in classroom instruction, whereas professional titles may, to some extent, indicate teachers' levels of professional expertise. In sum, the participants engaged in the formal assessment span a spectrum of varying professional levels within the teaching profession.

Instrument and Procedures

The instrument development process comprised four key steps: (1) developing a framework, (2) generating items, (3) reviewing items, and (4) pilot test and validation. Following the confirmation of reliability and validity through empirical evidence, a formal test was conducted to assess upper-secondary school biology teachers' PCK of scientific thinking.

The instrument's development was guided by two primary criteria. Firstly, it centered on crafting items grounded in authentic contexts. Acknowledging the implicit and dynamic nature of PCK, it was crucial to steer clear of overly simplistic scenarios that overlook the intricacies of teaching (Baxter & Lederman, 1999). Evaluating PCK entailed a concentration on teachers' capacity to navigate the distinctive and non-generalizable elements of the classroom setting (Kagan, 1990).

Secondly, a combination of multiple-choice and open-ended item formats was utilized to measure PCK. Multiple-choice items were chosen to enable assessment across broader samples and a wider spectrum of content areas (Bacon, 2003; Lipton & Huxham, 1970). However, it is recognized that the potential answers might be restricted by multiple-choice questions and could potentially be influenced by test-taking strategies (Hill et al., 2008). In contrast, open-ended items afford a chance to delve into teachers' unique teaching experiences without



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constraining responses to predefined choices. Nonetheless, this format necessitates additional effort and objective scoring (Schmelzing et al., 2013). Therefore, the inclusion of both item formats was deemed essential for comprehensively capturing PCK.

Developing a framework. One of the prevailing perspectives of PCK widely accepted by researchers is specificity to particular topics (Loughran et al., 2004; Park & Chen, 2012; Van Driel et al., 1998). Consistent with this notion, the PCK measure in this study was developed with a particular focus on scientific thinking, a key topic within the upper-secondary school biology curriculum. Drawing from established definitions in existing research (Grossman, 1990; Magnusson et al., 1999; Park et al., 2018; Shulman, 1986; Tamir, 1988), a framework for upper-secondary school biology teachers' PCK of scientific thinking was developed (Table 2).

Table 2

PCK Component	Description
Knowledge of eveningly (KO)	KC_A Teacher describes the connotation of scientific thinking and its elements
Knowledge of curriculum (KC)	KC_B Teacher identifies scientific thinking cultivated in the specific teaching process
Knowledge of students'	KSU_A Teacher knows students' difficulty in scientific thinking
understanding (KSU)	KSU_B Teacher enumerates and analyzes common ideas or answers of students in scientific thinking
Knowledge of instructional	KIS_A Teacher uses instructional strategies or teaching sequences that align with instructional objectives to cultivate students' scientific thinking
strategies (KIS)	KIS_B Teacher uses instructional strategies such as argumentation, modeling, socioscientific issues, and history of science to cultivate students' scientific thinking
Knowledge of assessment	KA_A Teacher chooses appropriate ways to assess students' scientific thinking, such as asking questions, paper- and-pencil tests, and observation in activities according to instructional objectives
(NA)	KA_B Teacher sets appropriate rating rubric to assess students' scientific thinking

Assessment Framework of PCK of Scientific Thinking

Generating items. In line with previous studies (Großschedl et al., 2019; McNeill et al., 2016; Park et al., 2018), the developed instrument encompasses a blend of multiple-choice and open-ended items. These items were created based on an examination of literature and classroom instructional videos pertinent to scientific thinking. Leveraging excerpts from teaching records and instructional designs as contextual materials, six vignettes were identified, covering topics such as (1) the characteristics of enzymes, (2) mitosis, (3) Mendel's experiments and laws of inheritance, (4) protein synthesis, (5) energy flow in ecosystems, and (6) genetically modified organisms. The initial version of the instrument comprised 20 items distributed across these six vignettes.

Reviewing items. The items underwent a rigorous review and selection process. A panel of twelve experts specializing in biology education was assembled, comprising two experts in biology education research, four seasoned upper-secondary school biology instructors, and five faculty members with teaching and research expertise. Three-quarters of the panelists had accumulated over a decade of teaching experience in this field. Each expert was assigned to assess the items regarding their clarity and adherence to the designated components. They were also encouraged to suggest revisions or justify the exclusion of items if they identified any issues with wording or content.

This methodical procedure was designed to enhance the instrument's content validity. Following the experts' review, it was found that 9 (45%) of the 20 items remained unchanged, 6 (30%) underwent modifications, 3 (15%) were deleted, and 1 new item was added. Notably, the vignette of Mendel's experiments and laws of inheritance was deleted due to the elimination of all the items within this context. Examples of the revised items are presented in Figure 1.



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Figure 1

Example of the Instrument's Item S2-1(KIS), S2-2-1 and S2-2-2(KA)

S2. During the lesson planning session, the teachers reached a consensus to incorporate the following five student activity steps into the teaching of **mitosis**.

① View animations of mitosis in plant and animal cells.

2 Observe mitosis under the microscope and describe the

characteristics of different cells.

③ Sort images of different stages of mitosis.

④ Simulate the behavior of chromosomes during mitosis using

various-colored pipe cleaners.

⁽⁵⁾ Construct a model of cell mitosis.

1. During the lesson preparation and discussion, two ideas about the lesson sequence emerged. Which one do you think is more conducive to developing students' scientific thinking? Please provide the reasons for your choice.

2. A teacher intends to employ the following two tasks as homework assignments to assess students' scientific thinking. Do you think they are viable?

1) Analyze the changes in the proportions of chromosomes and DNA molecules throughout cell mitosis.

2) The image below shows different regions of the root tip captured under a microscope. Based on the image, which of the following statements is incorrect?



A. To observe cells with the characteristics of the image (a), you should initially choose the field of view as shown in image (b), and subsequently magnify it for detailed observation.

B. Cell ② in image (a) is in the process of forming a cell plate, which is associated with the formation of the cell wall.

C. Most of the cells in image (c) are in interphase, while a few have undergone chromosome duplication, indicating the onset of mitosis.

Note: Images (a) - (c). From "Bianzhi zonghexing shiti tigao fuxi youxiaoxing [Compiling test questions to improve review effectiveness]," by Chen Y. Y., 2014, Shengwuxue Tongbao, 49(06), p. 63. (in Chinese). Copyright 1994-2023 by China Academic Journal Electronic Publishing House.



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Pilot test and validation. The pilot test serves four primary purposes: (1) to examine the appropriateness of item difficulty relative to the teacher's ability level, (2) to gauge the time needed for the teacher to complete the test, (3) to develop preliminary scoring rubrics by collecting typical responses from teachers, and (4) to evaluate the clarity of the instrument's phrasing, assess teachers' comprehension, and ensure that the items accurately represent the intended content. In the initial phase, a pilot test involving 47 upper-secondary school biology teachers, both pre-service and in-service, was conducted using convenience sampling. After completing the survey, some participants were selected to engage in discussions regarding their comprehension of the items and the reasoning behind their responses (Karabenick et al., 2007). The Rasch model was used to examine item quality. Combining interviews with Rasch analysis, 2 (11%) of the 18 items were removed as all participants answered them correctly, while 4 (22%) were modified due to low item discrimination. To ensure adequate item coverage for each dimension, 4 new items were added. Seven experts were invited to assess these new items' content validity. All 4 items received excellent ratings (S-CVI = 0.94).

To further refine the quality of the instrument, a second-round pilot test was conducted, employing convenience sampling to enlist 115 in-service upper-secondary school biology teachers. Subsequent to the second-round pilot test, 1 item was removed due to low item discrimination, and adjustments were made to 7 (35%) scoring rubrics following deliberation with experts. The final iteration of the instrument comprised 19 items. Table 3 illustrates the distribution of these items corresponding to various components of PCK related to scientific thinking. The complete instrument is presented in Appendix.

Table 3

Item Distribution Regarding PCK of Scientific Thinking

PCK Component	Items
Knowledge of curriculum (KC)	S1-4; S3-2; S4-1; S5-2; S5-3-1
Knowledge of students' understanding (KSU)	S1-1-1; S1-1-2; S4-2-1; S5-3-2
Knowledge of instructional strategies (KIS)	S1-2; S2-1; S3-1; S4-2-2; S5-3-4
Knowledge of assessment (KA)	S1-3; S2-2-1; S2-2-2; S5-1; S5-3-3

In science education, the Rasch model has been extensively utilized to refine and adapt instruments, leading to the creation of higher-quality items and measures (Boone & Scantlebury, 2006; Liu & Boone, 2023; Sideridis, 2007). Through the Rasch model, item and person invariance can be achieved, laying the groundwork for distinguishing between person and items (Engelhard & Wang, 2021).

Validation of the instrument was conducted employing the Rasch model. Rasch model is inclined to reveal more items for misfitting, thus providing extensive opportunities for enhancing item quality and generating invariant measures (Liu & Boone, 2023). Rasch measurement enables the establishment of a unified interval scale for both person ability and item difficulty, allowing for comparisons among respondents without requiring responses to all survey items (Boone et al., 2014; Boone & Scantlebury, 2006).

Formal test. An online examination was employed to assess the participants' PCK of scientific thinking following the validation of the instrument. Subsequently, the survey data underwent analysis utilizing the Rasch model.

Data Analysis

A multi-dimensional Rasch analysis was performed using the ConQuest software. Due to the items not sharing identical scale steps, a Rasch model for partial credit scoring was conducted. Person/item separation reliability was calculated to examine the instrument's reliability. The Expected A Posteriori/ Plausible Value (EAP/PV) reliability, which reflects the proportion between true and observed variance across each dimension (He et al., 2023), was computed to illustrate the precision degree to which the measurement of the constructs distinguishes the examinees (Wang & Lu, 2021). Unweighted/weighted fit statistics were calculated to evaluate the fitness between the data and the Rasch model. Additionally, The Wright map was employed to visually examine the instrument's validity. SPSS 26.0 was employed to analyze the teachers' PCK value generated through ConQuest.



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Research Results

Evidence for Reliability and Validity

Reliability. Reliability signifies the extent to which measurement outcomes can be replicated (Ding, 2023). The separation reliability of the instrument was determined to be .922. Furthermore, The EAP/PV reliability of each dimension was as follows: .808 (KC), .796 (KSU), .781 (KIS), and .726 (KA), all of which were deemed acceptable (Table 4).

Table 4

EAP/PV of Each Dimension

Dimension	PCK component	EAP/PV reliability
1	Knowledge of curriculum (KC)	.808
2	Knowledge of students' understanding (KSU)	.796
3	Knowledge of instructional strategies (KIS)	.781
4	Knowledge of assessment (KA)	.726

Validity. The Rasch fit statistics and Wright map serve as valuable tools for scrutinizing the validity of the computed measures (Bond & Fox, 2015). The unweighted and weighted statistics' mean-square (MNSQ) are derived through a chi-square analysis to assess the level of correlation, while the z-standardized (ZSTD) values represent *t*-test statistics indicating the likelihood of MNSQ occurrence by chance (Boone et al., 2014). Given the dependency of ZSTD values on MNSQ, the primary focus is placed on MNSQ for fit evaluation; provided that MNSQ falls within a reasonable range, the ZSTD value is not considered (Boone et al., 2015). According to Wright and Linacre (1994), the permissible spectrum of MNSQ values in the context of the survey usually falls between 0.6 and 1.4. As depicted in Table 5, unweighted MNSQ values for all items varied between 0.73 and 1.24, while weighted MNSQ values spanned from 0.83 to 1.37, indicating acceptable model-data-fit.

Table 5

Fit Statistics of Items

ltem	Estimate	Error	Unweighted MNSQ	Weighted MNSQ
S1-1-1	2.295	0.279	0.91	0.92
S1-1-2	1.415	0.345	0.84	0.89
S1-2	1.510	0.360	0.91	1.00
S1-3	0.445	0.216	0.97	1.03
S1-4	0.089	0.286	1.13	1.11
S2-1	1.442	0.192	1.19	1.37
S2-2-1	2.103	0.374	0.73	0.87
S2-2-2	1.967	0.362	0.94	0.97
S3-1	312	0.226	1.07	1.03
S3-2	1.069	0.228	0.82	0.89
S4-1	1.403	0.239	0.78	0.88
S4-2-1	2.402	0.304	1.24	1.18
S4-2-2	0.656	0.226	0.93	0.93
S5-1	0.259	0.159	1.13	1.10
S5-2	0.998	0.127	1.16	1.17
S5-3-1	842	0.155	1.04	1.02
S5-3-2	1.916	0.205	1.03	1.04
S5-3-3	1.587	0.178	0.76	0.83
S5-3-4	0.299	0.157	0.86	0.87

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The Wright map provides a visual depiction of person ability and item difficulty using a linear logit scale (Black et al., 2011). The difficulty level of the item is located on the map's right, whereas the ability of the person lies on the other side. The average value of item difficulties is set at 0 logits by default, with positive logits indicating probabilities above average and negative logits indicating probabilities below average (Bond & Fox, 2015). The Wright map facilitates the examination of whether the hierarchy of the items within a test aligns with the anticipated structure, thus enabling the evaluation of the instrument's construct validity (Boone & Staver, 2014). In the current study, item difficulty ranged from 0.842 to 2.402. As illustrated in Figure 2, item difficulty was appropriately dispersed, with thresholds covering the majority of participants' ability range at each difficulty level. This distribution suggests that the assessment instrument demonstrates good construct validity. Additionally, the Wright map highlights cases where individual ability falls below the item difficulty range, suggesting a requirement for simpler items.

Figure 2

Wright Map of The Thurstonian Thresholds for Each of The Items in The Second-Round Pilot Test

		Dimens	sion		Generalised-I	tem Thresho	lds	
	1	2	3	4				
6								
5					 S4-2-1. 2			
4					S5-3-2.2			
3		X X X X			S1-1-1.2 S5-3-3.2	S5-3-4.3		
2		X XX XXX XX	X XX XX	X XX	S2-1. 3 S5-2. 3 S2-2-1 S2-2-2	S3-2. 2	S4-1.2	
1	X XXX XXX XXXX	XXXX XXXX XXXX XXXX	XXX XXXX XXXX XXXXX	XX XXXX XXXX XXXXXXXXX	S2-1.2 S1-1-2 S1-1-1.1	S4-2-2.2 S1-2 S3-1.2	S5-3-1.3 S1-3.2 S5-2.2	S5-1.2
	XXXXXXXX XXXXXXXXXX XXXXXXXXXXXX XXXXXX	XXXXX XXXXX XXXX XXXX XXXX	XXXXXX XXXXXXX XXXXXXXXX XXXXXXXXX XXXX	XXXX XXXXXXXX XXXXX XXXXXXXXXXX	S4-1. 1 S5-3-3. 1 S1-4	S2-1.1	S3-2.1	S4-2-1
0	XXXXXXXXXXX XXXXXXXXXX XXXXXXXX XXXXXXX	XXXXX XXXX XXXX XXXX XXXX	XXXXX XXXXXX XXXXXX XXXXXX	XXXXX XXXXXXXX XXXXXXXX XXXXXXX XXXX	S5-3-2.1 S4-2-2.1 S1-3.1	S5-2.1	S5-3-4.2	
-1	XXXX XXX XXX XXX XXX XXX XXX	XXXX XXX XXX XXX XXX	XXXX XXXX XXX XXX XX	XXXX XXXXX XXX XXX XXX		S5-3-1.2		
-2	X	XXX XX XX XX	XXX XXX X	XX XX X	S5-3-4.1 S5-3-1			
-3		X X X						
-4		X						
-5								
-6								

Each 'X' represents $1.3\ \rm cases$ Dimension 1, Dimension 2, Dimension 3, and Dimension 4 are person ability estimates for KC, KSU, KIS, and KA, respectively



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PCK of Scientific Thinking Within the Participants

After calculating person ability using ConQuest, SPSS 26.0 was used to analyze the status of PCK of scientific thinking among the participants. Descriptive statistical data for each component of PCK of scientific thinking is displayed in Table 6. The mean ability estimates for upper-secondary school biology teachers across the four components varied as follows: 0.06 for KC, 0.58 for KSU, 0.18 for KIS, and 0.03 for KA. Notably, the mean ability of KSU is the highest, followed by KIS, KC, and KA.

Table 6

Descriptive Statistics of PCK of Scientific Thinking among Upper-Secondary School Biology Teachers

Component	Ν	Min.	Max.	М	SD
KC	292	-1.65	2.12	0.06	0.69
KSU	292	-1.67	3.51	0.58	0.99
KIS	292	-1.59	2.38	0.18	0.76
KA	292	-1.59	1.89	0.03	0.67

To examine the statistically significant differences in the abilities across the four components of PCK of scientific thinking, the Shapiro-Wilk test method (He & Wang, 2014) and paired samples *t*-test were calculated. Furthermore, Cohen's *d* was computed to gauge the effect size (Wen et al., 2016), with interpretations provided by Ferguson (2009) (minimum = 0.41, moderate = 1.15, strong = 2.70). The Shapiro-Wilk test did not show evidence of non-normality for KC (W = .995, p = .557), KSU (W = .994, p = .277) KIS (W = .995, p = .505) and KA (W = .996, p = .588).

As depicted in Table 7, the paired samples *t*-test's outcomes comparing teachers' abilities across the components revealed the following: Teachers' ability in KC (M = 0.06, SD = 0.69) was significantly lower than that of KSU (M = 0.58, SD = 0.99), t(292) = -19.656, p < .001, Cohen's d = 1.15. Teachers' ability in KC (M = 0.06, SD = 0.69) was significantly lower than that of KIS (M = 0.18, SD = 0.76), t(292) = -6.542, p < .001, Cohen's d = 0.38. There was no significant difference between the KC (M = 0.06, SD = 0.69) and KA (M = 0.03, SD = 0.67), t(292) = 1.367, p = .173. Teachers' ability in KSU (M = 0.58, SD = 0.99) was significantly higher than that of KIS (M = 0.18, SD = 0.76), t(292) = 24.939, p < .001, Cohen's d = 1.46. Teachers' ability in KSU (M = 0.58, SD = 0.99) was significantly higher than that of KI (M = 0.18, SD = 0.76), t(292) = 23.223, p < .001, Cohen's d = 1.36. Teachers' ability in KIS (M = 0.18, SD = 0.76) was significantly higher than that of KA (M = 0.03, SD = 0.67), t(292) = 23.223, p < .001, Cohen's d = 1.36. Teachers' ability in KIS (M = 0.18, SD = 0.76) was significantly higher than that of KA (M = 0.03, SD = 0.67), t(292) = 12.924, p < .001, Cohen's d = 0.76.

Moreover, an examination of the Wright map (Figure 2) revealed the overall distribution of teachers' ability in the KSU dimension surpassed that of other dimensions, while the overall distribution of teachers' ability in the KIS dimension was higher than that of KA.

Table 7

The Result of The Paired Samples T-Test

	М	SD	SEM	t	df	p	Cohen's d
KC - KSU	515	0.448	0.026	-19.656	291	< .001	1.15
KC - KIS	116	0.304	0.018	-6.542	291	< .001	0.38
KC - KA	0.031	0.391	0.023	1.367	291	.173	
KSU - KIS	0.399	0.273	0.016	24.939	291	< .001	1.46
KSU - KA	0.546	0.402	0.024	23.223	291	< .001	1.36
KIS - KA	0.148	0.195	0.011	12.964	291	< .001	0.76

From the specific responses of teachers in the paper-and-pencil test, the majority of teachers appeared to encounter challenges in comprehending the connotation of scientific thinking and its constituent elements within the realm of KC. Furthermore, they struggled to effectively identify scientific thinking cultivated in the specific teaching segments. For instance, item S3-2 (see Appendix) shows materials to help students understand RNA as



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a messenger molecule, and to examine whether teachers can recognize the scientific thinking elements and explain the reason. The result indicated that 48.97% of teachers successfully pointed out the element. Among them, only 11.03% provided further substantiated justifications. Conversely, 51.03% of teachers concentrated solely on the activities inherent to the given instructional context, thereby answering "information extraction" or "picture comprehension" rather than specific scientific thinking elements such as induction or deduction.

In the dimension of KSU, while most teachers can enumerate common student responses, ideas, or challenges in applying scientific thinking from a single perspective, they face difficulty in conducting comprehensive analyses of students' scientific thinking from multiple aspects. For example, item S4-2-1 (see Appendix) seeks to examine whether teachers can list difficulties students encounter in simplifying the Cedar Bog Lake food cycle and explain the reasons for these difficulties. Of the teachers surveyed, 53.06% can analyze from a singular perspective, while 6.12% can analyze difficulties and their reasons based on internal factors such as students' conceptual grasp and external factors such as the complexity of food web diagrams. However, 40.82% of teachers struggled to provide specific analyses of student difficulties in this context and offered generalized responses. Such as "students lack problem-solving skills" or "cannot fully comprehend figure information".

In the dimension of KIS, most teachers demonstrated a strong grasp of the applicability of various teaching strategies in fostering scientific thinking within teaching themes. However, they faced challenges in selecting appropriate teaching activities or sequences based on teaching objectives. For instance, in item S2-1 (see Appendix), teachers are assessed on their ability to select a teaching sequence more suitable for fostering students' scientific thinking in the teaching of mitosis. About 60.69% of teachers correctly chose the sequence that is more conducive to fostering students' scientific thinking. Further analysis of teachers' responses reveals that only 6.90% of them can comprehensively point out why idea 1 is more helpful in fostering students' scientific thinking compared to idea 2. Additionally, 39.31% of teachers chose idea 2, these teachers were more concerned with students' learning interests and the ease of knowledge acquisition when selecting teaching sequences.

In the dimension of KA, most teachers struggled to select appropriate approaches to assess students' scientific thinking and to establish suitable grading criteria for assessing their scientific thinking. For example, item S5-3-3 (see Appendix) evaluates teachers' ability to set rubrics for assessing different levels of student scientific argumentation. From the responses, only 4.11% of teachers can comprehensively establish grading criteria based on both the elements and quality of argumentation. 38.01% of teachers can set rubrics from one aspect, indicating that these teachers may understand aspects of high-quality argumentation but still need to enhance their understanding of the dimensional and grading criteria of argumentation. About 57.88% of teachers used "excellent, good, poor" or "high, medium, low" to describe grading criteria, and did not give any specific performance related to argumentation.

Discussion

The development of the instrument in this research is based on the Rasch measurement method. It is widely regarded as an advanced approach to measurement and is extensively applied in science education, particularly in the validation and refinement of instruments (Boone et al., 2010; Liu & Boone, 2023; Sondergeld & Johnson, 2014), which enables researchers to utilize respondents' raw test or scale scores and represent their performance on a linear scale, accommodating the varying difficulties of all test items (Boone, 2016). This addresses limitations inherent in traditional Classical Test Theory (CTT), such as the interdependence between item difficulty and person ability, challenges in conducting equating comparisons, and susceptibility of measurement parameters to sample quality (Sondergeld & Johnson, 2014). In this study, the Rasch model offered various evidence to help improve the quality of items, such as EAP/PV reliability, fit statistics like unweighted and weighted MNSQ, Wright maps, etc.

This confirms what Liu and Boone (2023) said that the Rasch model is helpful in creating more opportunities for improving items and generating invariant measurements. Assessing PCK is challenging (Park et al., 2018). Chan and Hume (2019) noted a lack of effective measurement tools for investigating teachers' PCK in authentic classroom settings on a large scale. This study addresses this gap by simulating authentic classroom contexts and employing the Rasch model to design a quantitative instrument. Furthermore, while some research analyzes overall PCK scores or component integration (e.g. Jüttner et al., 2013; Park & Chen, 2012; Schiering et al., 2022), they struggle to assess individual PCK components' quality. As the PCK rubric can overcome this problem (Chan et al., 2019), this study uses the Rasch model to obtain ability values for each component of PCK of scientific thinking, which will contribute to further understanding the impact of individual components' quality on overall PCK performance. Building upon this foundation, future research needs to consider how to simultaneously reflect the quality of each component of PCK and its interconnection and integration.



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The research results reflect the performance of teachers across the four components of the PCK of scientific thinking. In terms of KUS, Park et al. (2020) found that in-service teachers perform well when asked to identify students' misconceptions or learning difficulties in given scenarios. However, their performance appears inferior when prompted to provide reasoning or explanations for why students often encounter misconceptions or difficulties. Similar results were observed among teachers in this study. The performance of teachers on KIS varies across different studies. Some findings suggest that teachers may struggle to elucidate how to implement these teaching strategies in their instruction (Bektas et al., 2013) or to explain why their chosen teaching strategies effectively address student difficulties or misconceptions (Park et al., 2020). Conversely, another study indicates that teachers demonstrate strong performance in terms of instructional strategies (Hanuscin et al., 2010). In terms of KC, the findings from this study mirror those of earlier research (Akinoglu & Dilek, 2015), suggesting a deficiency in teachers' comprehension of scientific thinking. This may be attributed to the backdrop of curriculum reform, where the enactment of the updated version of the Upper-Secondary School Biology Curriculum Standards (MEPRC) in 2020 explicitly emphasizes scientific thinking as an element of students' key competencies. This indicates that scientific thinking represents a novel concept for Chinese upper-secondary school biology teachers. As for KA, the performance of the teachers is relatively poor. This consistent pattern has been documented in prior research (Hanuscin et al., 2010). Some researchers deem teachers' understanding of assessment superficial, lacking clearly defined objectives, and predominantly reliant on personal teaching experiences (Wang, 2008; Yang, 2009). Furthermore, some research has found that even after participating in some professional development programs, teacher performance on KA still did not improve (Bektas et al., 2013).

The results suggest that researchers should pay particular attention to the development of teachers' PCK of scientific thinking in the areas of KC and KA. Given the constrained access of teachers to structured professional training, it is crucial to develop targeted programs focused on addressing teachers' needs and weaknesses (Grierson & Woloshyn, 2013). These programs ought to elucidate the definition, connotation, and relationship of scientific thinking and its constituent components, which will assist teachers in developing a systematic understanding of scientific thinking. Additionally, professional development programs should strive to assist teachers in forging a coherent link between instructional objectives and assessment approaches while also tackling challenges related to identifying appropriate dimensions for assessing students' scientific thinking.

Conclusions and Implications

Given the lack of previous research measuring teachers' PCK of scientific thinking, this study constructed an instrument specifically tailored for PCK of scientific thinking among upper-secondary school biology educators. The validation procedure during instrument development included content validity, assessed through expert review and participant interviews, as well as construct validity, evaluated through analysis of MNSQ and the Wright map. The analysis results of the Rasch model indicate that the instrument's overall separation reliability is greater than 0.9, and the reliability of EAP/PV for each dimension exceeds 0.7 (see Table 4). The fit statistics for MNSQ are all within reasonable ranges (see Table 5), and the Wright map shows that the difficulty of the items covers the ability values of most individuals (see Figure 2). This evidence collectively indicates that the instrument demonstrated overall high quality. Nonetheless, there remain areas for enhancement. Specifically, the overall distribution of item difficulty slightly exceeded the range of person ability, indicating a requirement for additional easier items or adjustments to mitigate the difficulty of certain items.

From the perspective of the teacher's PCK of scientific thinking assessment results, the overall performance tends to fall within the middle to lower levels. Only a small proportion of teachers demonstrate proficiency at a higher level, highlighting the imperative for enhancing training in scientific thinking. Across the four constituent components comprising PCK of scientific thinking, performance levels are delineated in descending order as follows: knowledge of students (KSU), knowledge of instructional strategies (KIS), knowledge of curriculum (KC), and knowledge of assessment (KA).

Given that this study adopts a cross-sectional design, focusing on the current status of PCK of scientific thinking among upper-secondary school biology teachers, future longitudinal research endeavors could explore the development trajectory of PCK of scientific thinking across different stages and investigate the key factors influencing its evolution. Furthermore, future research could utilize the instrument developed in this study to classify teachers into different proficiency levels by describing the specific performance associated with each level. These would provide additional evidence and bolster support for the development of targeted professional development courses.

Several limitations need to be acknowledged. Firstly, there is a recognized need for additional items to be



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added to the instrument to enhance its comprehensiveness. Secondly, despite our intention to develop an instrument aimed at measuring the status of PCK of scientific thinking among Chinese upper-secondary school biology teachers, the sample obtained lacks sufficient regional balance nationwide due to constraints in resources and support. Thirdly, the instrument was developed in Chinese, necessitating validation in other cultural and linguistic contexts if translated for use.

Declaration of Interest

The authors declare no competing interest.

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Appendix

PCK of Scientific Thinking Assessment

Dear teacher:

Welcome to the questionnaire designed to explore the present state of upper-secondary school biology teachers' pedagogical content knowledge (PCK) of scientific thinking. Your participation in this survey is greatly appreciated. Please rest assured that your responses will remain anonymous and confidential, solely for research purposes. Kindly complete the questionnaire thoughtfully, drawing upon your understanding and expertise.

We sincerely thank you for your invaluable support and cooperation.

Basic Information

1. Your gender:

□ Male □ Female

2. What is your highest level of educational attainment? (for pre-service teachers, please select the highest level of education you are currently pursuing):

□ Below Bachelor's degree □ Bachelor's degree □ Master's degree □ Ph. D.

3. What is your major field of study for your highest academic degree?

□ Biology-related field (such as biological science, biotechnology, botany, zoology, ecology, molecular biology, biochemistry, etc.)

□ Education-related field (such as Biology Education, Curriculum and Instruction in Biology, Subject Teaching in Biology, Science Education, etc.)

🗆 Other ____

- 4. Your years of teaching experience are _____
- 5. Your professional title:
 - Undetermined Rank Entry Level
 - □ Intermediate Level □ Deputy Director Level and Above
- 6. Your province of employment: _____
- 7. What type of school do you work at?
 - National Demonstration School
 Provincial Demonstration School
 - Municipal Demonstration School
 District/County-level Demonstration School
 - Regular Secondary School

Below are vignettes related to scientific thinking in upper-secondary school biology, along with corresponding questions. Please read the questions carefully and respond as instructed. Thank you for your cooperation!

Note: Multiple-choice questions in the questionnaire may have one or more correct answers.

S1. In the teaching of **enzyme characteristics**, one of the objectives is to cultivate student's ability to models and modeling.

1. Within the classroom, the teacher employs the experimental demonstration (see Figure a) to foster students' intuitive understanding of how pH influences protein properties, thereby guiding their speculation and description of pH's impact on enzyme activity (see Figure b). Please analyze the possible idea or basis for students' drawing of curves ① and ②.



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1) Possible idea or basis for students' drawing of **curves** ①:

2) Possible idea or basis for students' drawing of **curves** 2):

2. After organizing the experiment on factors influencing enzyme activity and guiding students in data processing, a group of students produced the following curve. To foster students' scientific thinking in models and modeling, the **most** appropriate next step in the teaching process would be:



The effect of pH on catalase acticity

A. Present the curve graph from the textbook and guide students to compare and analyze similarities and differences.

B. Organize student discussions to recognize the need to collect more data by narrowing the pH concentration gradient.

C. Guide students to deduce how pH affects catalase activity based on the graph and determine the optimal pH.

D. Guide students to attempt drawing a curve graph illustrating the effect of temperature on catalase activity using the experimental results.



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3. To evaluate students' models and modeling abilities in this class, which of the following three assessment tasks is the **most** effective:

A. Evaluation through the following classroom questioning

① Which enzyme would be most appropriate for experimentation, and why?

2 What are the independent and dependent variables in the experiment?

③ How to control the independent variables? How to observe or detect the dependent variables?

B. Evaluation through the following classroom activities

① Design a table to record experimental results.

2 Plotting the curve of the relationship between pH and enzyme activity based on experimental results.

③ Analyzing the strengths and weaknesses of the findings reported by other groups

C. Evaluation through the following in-class exercises

The figure below represents a model of enzyme action. Try to describe this model in words. What characteristic(s) of enzymes can this model explain?



4. Among the following concepts of the curriculum standard, which one is **not** suitable for cultivating students' ability in models and modeling:

A. Describing that cells are all enveloped by a plasma membrane, which separates the cell from its environment, controls the passage of substances in and out, and participates in intercellular communication.

B. Describing that base substitutions, insertions, or deletions can alter the base sequence in a gene.

C. Providing examples to illustrate that immune cells, immune organs, and immune-active substances constitute the structural and material basis of immune regulation.

D. Elaborate on how gene segregation and independent assortment during sexual reproduction result in multiple possible genotypes and phenotypes in offspring, allowing for the prediction of hereditary traits in offspring

S2. During the lesson planning session, the teachers reached a consensus to incorporate the following five student activity steps into the teaching of **mitosis**.

2 Observe mitosis under the microscope and describe the characteristics of different cells.

③ Sort images of different stages of mitosis.

- $({ 4 \hspace{-.15cm} I \hspace{-.15cm} })$ Simulate the behavior of chromosomes during mitosis using various-colored
- pipe cleaners. ⑤ Construct a model of cell mitosis.

1. During the lesson preparation and discussion, two ideas about the lesson sequence emerged. Which one do you think is more conducive to developing students' scientific thinking? Please provide the reasons for your choice.



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2. A teacher intends to employ the following two tasks as homework assignments to assess students' scientific thinking. Do you think they are viable?

1) Analyze the changes in the proportions of chromosomes and DNA molecules throughout cell mitosis.

2) The image below shows different regions of the root tip captured under a microscope. Based on the image, which of the following statements is incorrect?



A. To observe cells with the characteristics of the image (a), you should initially choose the field of view as shown in image (b), and subsequently magnify it for detailed observation.

B. Cell ② in image (a) is in the process of forming a cell plate, which is associated with the formation of the cell wall.

C. Most of the cells in image (c) are in interphase, while a few have undergone chromosome duplication, indicating the onset of mitosis.

S3. In the teaching of **genes guiding protein synthesis**, the teacher asks, "How do genes interact with ribosomes?" and guides students to discuss and make hypotheses.

1. After students propose hypotheses, which of the following approaches is **most** consistent with your perspective:

- A. Guide students to conduct experiments to obtain evidence to verify the validity of their hypotheses.
- B. Present a video on DNA transcription, guiding students in analyzing it and drawing conclusions.

C. Guide the data analysis on ribosomes, chromatin, and other materials to assess the validity of the hypotheses.

D. Present scientists' research findings to students and then explain the specific processes.

E. If you have another approach, please add _____



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2. The teacher presents the following materials to help students understand RNA as a messenger molecule.

Material 1	Material 2	Material 3
Incubate salivary gland cells of <i>Drosophila hydei</i> in a solution con- taining ³ H- Uridine, then observe the chromosomes.	Rich carried out the first DNA- RNA hybridiza- tion and made a hybrid double helix.	Barbara J. Stevens observed salivary gland cells of <i>Chironomus thummi</i> under an electron microscope and found that granules ranging from 400 to 500 A entered the cytoplasm from the nucleus through nuclear pores. Upon treatment with ribonuclease, these granules disappeared.
- ³ H-Uridine		

What kind of scientific thinking do teachers aim to cultivate in students through this teaching process? Briefly explain the reasons.

S4. Based on the content of energy flow in ecosystems, answer the following questions.

1. The teacher displays pictures of forests, grasslands, deserts, tundra, rivers, lakes, and oceans, posing the question, "Which type of ecosystem is more conducive to studying the energy flow in ecosystems?" What kind of scientific thinking do students mainly need to apply to answer this question? Briefly explain the reasons.

2. In the qualitative analysis of energy flow in Cedar Bog Lake, teachers found that a majority of students struggled with transforming Figure a into Figure b.





Figure a: A food cycle in Cedar Bog Lake



1) What difficulties might students encounter during the transition process? What could be the possible reasons?

2) Choose a potential difficulty and briefly explain how to overcome this teaching challenge.



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S5. Based on the content of the safety of genetically modified products, answer the questions.

1. One of the objectives of this lesson is to cultivate students' scientific argumentation. Which is the most effective way for teachers to understand the scientific argumentation level of the whole class before the lesson?

A. Conducting a debate on the safety of genetically modified foods.

B. Require students to search materials before class and outline the production process of genetically modified foods.

C. Require students to write a short essay on the topic of "Are genetically modified foods safe?"

D. If you have another approach, please add _

2. Regarding the question of "Are genetically modified foods safe?", the teacher presented three research results in class and asked students to assess the credibility of these results.

	Research 1	
	Chicken fed with	Chicken fed with
	non-genetically	genetically
	modified corn	modified corn
Live weight (kg)	1.6	1.6
Egg production	43.8	41.9

Note: there was no significant difference

Research 2

	Male mice fed with	Male mice fed
	non-genetically	with genetically
	modified corn	modified corn
Sperm number (10 ⁸ /g)	260.96	263.63
Abnormal rate (%)	1.97	2.00

Note: there was no significant difference

					1	Research 3									
	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
kD								_							
95								=							
72	-							-	1	1	11		-		_
55 43								=		17	53				
34						- Hand	-	-							-
26								-					-		-
															-
								-							

Digestive stability from recombinant 1Ab protein of genetically modified corn in simulated gastric and intestinal fluid

Note: 1, 9: 1Ab protein; 2-6: 1Ab protein digested by simulated gastric fluid for 0, 15s, 2, 30, 60min, respectively; 7: Gastric protease; 8: Marker; 10-14: 1Ab protein digested by simulated intestinal fluid for 0, 15s, 2, 30, 60min, respectively; 15: Pancreatic protease.

Claim : Ranking the Credibility of Evidence: ____ < ___ < ____

What kind of scientific thinking do teachers aim to cultivate in students through this teaching process? Briefly explain the reasons.

3. During group discussions based on the three research results provided by the teacher, Student A suggested, "I think the current genetic modification technology is not mature enough, so the safety of genetically modified foods still needs to be observed." On the other hand, Student B believes that genetically modified foods are safe. The viewpoint is as follows:

Student B: Consuming genetically modified foods is safe because studies have shown that the 1Ab protein of genetically modified corn degrades completely within 15 seconds to 2 minutes in simulated gastrointestinal fluids.

1) What scientific argumentation elements are contained in Student B's viewpoint?

A. Claim B. Evidence C. Reasoning

2) Based on your understanding of the students, apart from the perspectives of Student A and Student B, how else might students articulate their viewpoints? Please list as many different types of viewpoints that other students might propose as possible.



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3) Regarding student responses, establish grading criteria based on the level of students' scientific thinking.

4) What other contents are suitable for argument-driven inquiry in high school biology? Please provide at least two specific examples to illustrate.

Note.

- 1. The images (a) (c) in item S2-2-2. From "Bianzhi zonghexing shiti tigao fuxi youxiaoxing [Compiling test questions to improve review effectiveness]," by Chen Y. Y., 2014, *Shengwuxue Tongbao, 49*(06), p. 63. (in Chinese). Copyright 1994-2023 by China Academic Journal Electronic Publishing House.
- 2. The picture in material 1 in item S3-2. Adapted from "In situ transcription analysis of chromatin template activity of the X-chromosome of *Drosophila* following high molar NaCl treatment," by Chatterjee, R. N., Dube, D. K. & Mukherjee, A. S., 1981, *Chromosoma, 82*(4), p. 518. https://doi.org/10.1007/BF00295010. Copyright 1981 by Springer-Verlag.
- 3. The picture in material 3 in item S3-2. Adapted from "RNA transport from nucleus to cytoplasm in chironomus salivary glands," by Stevens, B. J., 1966, *The Journal of Cell Biology*, *31*(1), p. 71, 75. CC-BY-NC.
- 4. The figure a in item S4-2. Adapted from "The Trophic-Dynamic Aspect of Ecology," by Lindeman, R. L., 1942, *Ecology, 23*(4), p. 401. https://doi.org/10.2307/1930126. Copyright 1942 by the Ecological Society of America.
- 5. The figure b in item S4-2. Adapted from *Top 7 Characteristics of energy flow in an ecosystem*, by Meghna, G., 2016, (https://www.notesonzoology.com/ecosystem/top-7-characteristics-of-energy-flow-in-an-ecosystem-zoology/4212). Copyright by NotesOnZoology.com.
- 6. The figure in research 3 in S5-2. From "Zhuan G10evo he Cry1Ab/Cry2Ab jiyin yumi GAB-3 waiyuan jiyin biaoda danbai de xiaohua wendingxing [Digestive stability of recombinant G10evo, Cry1Ab/Cry2Ab proteins of transgenic corn GAB-3 in simulated gastric and intestinal fluid]," by Li, L., Wang, J. Zhao, Y. & Liu, H. L., 2015, *Huanjing yu Jiankang Zazhi, 32*(2), p.114. (in Chinese). Copyright 1994-2023 by China Academic Journal Electronic Publishing House.

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