ORIGINAL ARTICLE

Development of An Achievement Test on Organic Substances Within the Scope of the 9th-Grade Biology Course

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***Abstract**: This study aims to develop a valid and reliable achievement test in accordance with the content framework of the 9th-grade Biology Course Curriculum published within the scope of the Turkish Century Maarif Model on the subject of "Organic Matter". The screening method was used for this purpose. The sample of the study consists of 258 9th-grade students studying in two state schools located in the eastern part of Turkey. While creating the test, the following steps were followed: determining the purpose and scope of the test, literature review, writing and arranging the items, performing the preliminary application and analyzing the items, and finally creating the test. Initially, a draft test consisting of 60 questions was prepared. However, as a result of the item analyses, some items were eliminated in order to increase the validity and reliability of the test. At the end of this process, a 40-question achievement test that better reflects the scope of the curriculum and has high content validity was created. As a result, the achievement test, which consists of a total of 40 questions covering the curriculum content framework with an average difficulty of 0.33, an average discrimination index of 0.32 and a KR-20 reliability coefficient of 0.84, has taken its final form.*

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***Ethical Statement**; All study participants were provided with an informed consent form before the study began. Participants were clearly informed that participation in the study was entirely voluntary and that they could withdraw from the study at any time. Data obtained throughout the data collection process was anonymized; no personal or institutional information was collected or used. This study was approved by the Kafkas University Social and Human Sciences Scientific Research and Publication Ethics Board (No. 66 and No. 33, dated February 19, 2025).*

***Consent to Participate**: Informed consent was obtained from all individual participants included in the study. The process of obtaining informed consent was overseen and approved by the Social Sciences and Humanities Ethics Committee of Kafkas University, ensuring the study was conducted in accordance with the participants' consent.*

***AI Declaration**: The authors affirm that artificial intelligence did not contribute to the process of preparing the work.*

Introduction

EDUCATION is a multidimensional process that is not limited to the transfer of knowledge; it also reveals the extent to which this knowledge has been learned and how effective the teaching process is (Can, 2014). Decisions regarding teaching programs can only be made soundly with measurement and evaluation practices, which are an integral part of this process. In particular, determining the academic success levels of students is one of the most effective ways to understand the extent to which they have learned the given information (Ashraf & Zolfaghari, 2018; Bingöl & Halisdemir, 2024; Wafubwa & Csíkos, 2022).

The evaluation process is not limited to measuring student success; it also provides feedback to departments, schools or administrative units about the teaching process, curriculum and the quality of educational services, enabling continuous improvement in these areas (Kizlik, 2012). In this context, teachers working at all levels of the education-training process prepare and apply many tests throughout the year. Therefore, tests are seen as an indispensable tool in educational institutions (Quaigrain & Arhin, 2017). Especially at the end of the term, tests prepared to comprehensively measure students' knowledge levels provide teachers with valuable feedback not only about students' success levels but also on their own teaching activities. Therefore, the quality and reliability of tests is of great importance (Quaigrain & Arhin, 2017).

Academic success is generally associated with the student's performance in in-class and out-of-class activities, grades, and honors; therefore, academic evaluations play a central role in determining the quality of education (Kaur & Singh, 2022). Similarly, student evaluations of the teaching process have become one of the widely used methods to measure the quality of teaching in the academic world (Kreitzer & Sweet-Cushman, 2022).

Exams have an important place among the measurement tools used in this process. These tools, which are organized in various formats such as fill-in-the-blank, short answer, true-false, matching, open-ended and multiple-choice questions, are used to determine the extent to which students have achieved the specified gains (Balcı & Tekkaya, 2000). Although each type of test has its own strengths and weaknesses, according to research, the most commonly used measurement tool after interviews in revealing students' knowledge on a particular subject is multiple-choice tests (Ogan Bekiroğlu, 2004; Pektaş & Karamustafaoğlu, 2024). Although they are limited in measuring critical thinking and creativity, they are widely preferred because they are effective in determining misconceptions and academic success (Demir et al., 2016; Haladyna, 1997; Küçükahmet, 2002). In addition to the ease of application, the fact that the scoring is based on objective criteria

increases the reliability of the test and makes the evaluation process more systematic. All these features are among the main reasons why multiple-choice tests are widely used in educational environments (Kargın & Gül, 2021). For this reason, there are many studies and applications in both national and international literature on the development of achievement tests containing multiple-choice questions (Akçay & Önal, 2023; Ayvacı & Durmuş, 2016; Bolat, 2019; Çelik & Bayrak, 2025; Hurt & Mishra, 1970; Mamolo, 2021; Pandra & Mardapi, 2017; Saraç, 2018; Winarti et al., 2021). When the studies are examined, it is seen that the studies are especially widespread in the field of science education. In this field, biology is one of the basic courses that play an important role in students' understanding of the nature they live in, the relationships between living things in nature, their own body structures, their nutrition, and what they need to pay attention to in order to live a healthy life (Köse & Gül, 2016). In fact, in the literature, achievement test development studies on various biology topics such as cell division and heredity, respiration and photosynthesis, biological diversity, genetics, and systems are frequently encountered (Aydın & Selvi, 2020; Çakır & Aldemir, 2011; Kargın & Gül, 2021; Kaya & Gül, 2020; Kızkapan & Bektaş, 2018, Tekkaya & Balcı, 2003). However, achievement test development studies on the subject of Basic Components of Living Things are very limited (Elmacı, 2010; Şakir 2013; Şems, 2006), and these studies are achievement tests that include all inorganic and organic compounds. On the other hand, a multiple-choice test that covers only the subject of "Organic Substances", which constitute the basic building blocks of living things, on a broader scale and sufficiently evaluates it in terms of validity and reliability has not been encountered.

In the Biology Course Curriculum published by the Ministry of National Education (MoNE, 2024) within the scope of the Turkish Century Education Model, a comprehensive content framework has been presented regarding the subject of "Organic Substances". The subject of organic substances forms the basis of the concepts to be learned within the scope of the 9th grade biology course; it is one of the most important subjects in biology, especially since it is a preliminary information for understanding advanced units such as protein synthesis, systems and heredity (Sinan et al., 2006). Therefore, it is of great importance to measure the knowledge levels of students on this subject accurately and reliably, and at the same time, to prepare them with an inclusive approach that is suitable for the Education Model.

This study aims to develop a valid and reliable achievement test in accordance with the content framework of the 9th Grade Biology Course Curriculum published within the scope of the Turkish Century Education Model on the subject of "Organic Matter". In this context, the main question of the research was determined as follows: Is the achievement test developed

to determine the achievement levels of students on the subject of "Organic Matter" valid and reliable?

Method

This research was conducted using the survey method. The survey method is a research method that is usually conducted on larger samples. This method allows an in-depth examination of the participants' interests, skills, opinions, and talents regarding a topic or event (Fraenkel & Wallen, 2006). In the research, an achievement test was prepared to measure what the students learned about organic matter in the 9th-grade Biology course.

Study Group

The sample of the study consists of 258 9th-grade students (132 female, 126 male) studying in two public high schools located in the eastern region of Turkey. The students' ages ranged from 15 to 16 years. Additionally, for the reliability analysis of the finalized version of the achievement test, a separate sample of 204 9th-grade students (105 female, 99 male) from two other public high schools in the same region was used.

When the literature is examined, Maccallum (1999) recommends the sample size to be between 100 and 200; Çapık et al. (2018) emphasize that the sample size should be at least 200 in order to obtain valid and reliable results. Comrey and Lee (1992) considered a sample size of 200 as appropriate. Field (2018) states that the sample size should be at least five times the number of items depending on the number of items of the test. Accordingly, considering the number of items in the created test, the pilot study was conducted with 258 students and the reliability analysis with 204 students.

Pilot Testing and Data Collection Process

The pilot test was administered to 258 students in a classroom setting using printed test paper, and the average time for students to complete the test was determined to be 40 minutes. During the test administration, feedback was received from students regarding any technical or comprehension issues, and minor language adjustments were made to some items based on this feedback. Informed consent was obtained from the participants, and the study was approved by the ethics committee for ethical compliance.

Development Process of the Achievement Test on Organic Matter

The development process of the achievement test was carried out based on the test development steps suggested in the literature. The process followed in this study was structured similarly to the test development stages suggested by Haladyna (1997) and Kızkapan and Bektaş (2018). Accordingly, the stages followed and the procedures performed for the development of the achievement test are explained below:

Determining the Purpose and Scope of the Test

The main purpose of the achievement test developed in this study is to measure the academic success levels of students in the subject of "Organic Matter" in the 9th-grade Biology course. A comprehensive content framework has been presented in the Biology Course Curriculum published by the Ministry of National Education (MoNE, 2024) within the scope of the Turkish Century Education Model. The main topics covered in this framework are as follows:

- *Carbohydrates:*
  Monosaccharides: Ribose, Deoxyribose, Fructose, Glucose, Galactose
  Disaccharides: Sucrose, Maltose, Lactose.
  Polysaccharides: Glycogen, Starch, Cellulose, Chitin.
- *Lipids:*
  Fatty acids, triglycerides, phospholipids, and steroids.
- *Proteins:*
  Structure of amino acids, enzymes (simple and compound enzymes, activation energy, enzyme-substrate relationship), and factors affecting enzymatic reactions.
- *Nucleic Acids:*
  Structure of DNA and RNA.
- *Vitamins:*
  Fat-soluble vitamins and water-soluble vitamins.
  The test items developed in line with this scope have been carefully structured to cover the relevant contents and have been prepared to measure students' knowledge levels on these subjects in a valid and reliable manner.

Literature Review and Determination of Test Questions

The scope of the test to be developed was determined by examining the 9th-grade Biology Course Curriculum (MoNE, 2024) and then a comprehensive literature review was conducted by the researchers. In this process, previously developed achievement tests, especially on the subject of "Organic Matter" at the 9th-grade level, were carefully examined. As a result of the literature review, achievement tests created by various researchers

were found. For example, Şakir (2013) developed the Basic Compounds of Living Organisms Achievement Test in his study. This test consisted of 25 multiple-choice questions and was applied to 116 9th-grade students studying in different schools. According to the application results, the reliability coefficient of the test was calculated as Cronbach Alpha 0.73. Elmacı (2010) prepared 38 multiple-choice questions for the subject of Basic Components of Living Organisms and created an academic achievement test containing 20 of these questions. The reliability coefficient of the test was found to be Cronbach's Alpha 0.68, difficulty index 0.42 and discrimination index 0.44. Şems (2006) developed an achievement test on the subject of Basic Components of Living Things. This developed test consists of 55 multiple-choice questions and was applied to a total of 100 students studying in the second year of high school. The reliability of the test was calculated as Cronbach's Alpha coefficient = 0.91, which shows that the test is highly reliable.

As a result, when the studies conducted are examined, the purpose of developing a new achievement test in the current study is the need for an up-to-date, comprehensive and pedagogically appropriate assessment tool to measure students' academic achievements in the subject of "Organic Matter" in the 9th-grade Biology course in a valid and reliable manner. When the achievement tests developed by different researchers in the literature are examined, it is seen that these tests attract attention with their different content structures, application samples and reliability levels. However, due to reasons such as the fact that most of these tests have a limited number of questions and do not fully overlap with the current curriculum, a new achievement test was needed.

Achievement Test Development Process: Draft Preparation and Expert Opinions

In the study, by examining the relevant literature, an "Organic Substances Achievement Test" (OSAT) was prepared, which includes 60 multiple-choice questions, each consisting of five options in accordance with the content framework regarding the subject of "Organic Substances" in the 9th-grade Biology Course Curriculum published within the scope of the Turkish Century Maarif Model. In order to ensure validity in scientific research, it is a common and accepted practice to seek the opinions of experts in the field (Aydın & Selvi, 2020; Gönen et al., 2011; İlhan & Hoşgören, 2017; Şen & Eryılmaz, 2011). In this regard, the draft form of the achievement test consisting of 60 multiple-choice items with five options was presented to the evaluation of three academicians competent in field knowledge and measurement and evaluation and one experienced biology teacher. The experts were given a structured written evaluation form to assess the items

based on criteria such as language clarity, content adequacy, scope validity, and visual presentation. They provided detailed written feedback on each item.

Following this, a face-to-face meeting was held with the experts to discuss any conflicting feedback and to reach consensus on necessary revisions. As a result of this collaborative process, several items were revised in terms of wording and clarity, some were reorganized to better align with the curriculum objectives, and others were removed due to redundancy or ambiguity. All modifications were documented thoroughly, and a consensus report summarizing the expert evaluations and the changes made was prepared. The final test form was created based on this rigorous expert review process.

In designing the multiple-choice items, each item consists of a clear and concise stem followed by four distractors and one correct option. The length and wording of the stems were kept consistent and age-appropriate, avoiding unnecessary complexity. Distractors were designed to reflect common student misconceptions or plausible alternatives to increase the discriminatory power of the items. Care was taken to ensure that the distractors were grammatically consistent with the stem and not obviously incorrect. Additionally, technical item-writing flaws such as the use of "all of the above," "none of the above," or unequal option lengths were strictly avoided. Items were revised after expert feedback to ensure content validity and clarity.

Item Analysis Procedures

Item Difficulty Index

The item difficulty index (Pj) shows the rate of correct answers for each item in the test, and this rate takes a value between "0" and "1". A value close to zero can be interpreted as the item being difficult, and a value close to one can be interpreted as the item being easy. In item analysis, the difficulty index values are required for the items in the test to be between 0.20 and 0.80 (Kubiszyn & Borich, 2003). The item difficulty indexes of the items were calculated to determine the difficulty level of each item in the test.

Item Discrimination Index

Item discrimination index (rjx) is a ratio used to distinguish between students in the lower and upper groups and can take a value between -1 and +1. An item discrimination index of zero indicates that the item is not discriminatory (Bayrak çeken, 2012). In studies, it is generally seen that items with an item discrimination index value of 0.19 and below should be removed from the

test, items between 0.20-0.29 can be corrected or used in cases of necessity, items between 0.30-0.39 are considered quite good items, and items with a value of 0.40 and above are considered very good items (Crocker & Algina, 2006).

Item Analysis Based on Subgroup-Upper Group Mean Difference

For the construct validity of OSAT, item analysis based on 27% subgroup-upper group mean difference was performed (Özçelik, 2010). In this item analysis, it was examined whether the difference between the two group means was significant or not, and an independent samples t-test was applied.

Correlation-Based Item Analysis

Correlation-based item analysis was applied to reveal the relationship between each item scores and the total score in OSAT (Büyüköztürk, 2010). In this analysis, the correlation (item-total correlation) value between each item and the scale scores (except the item in question) were calculated. In this analysis, items with an item-total correlation value above 0.20 were evaluated as good. Although some limit values were taken as criteria in the interpretation of the item-total score, Tavşancıl (2002) stated that items with a lower limit of 0.20 could be included if it was preferred to keep the item in the measurement tool.

Internal Consistency Reliability

After the item analyses, the Kuder-Richardson-20 (KR-20) formula was used to determine the internal consistency of the scale.

Validity Analysis

In addition to content validity, which was established through expert opinions, the validity of the test items was also supported through item analysis methods. In this context, item-total correlations, which reveal the relationship between each item and the total test score, were calculated. Values above 0.20 indicated that the relevant items made a significant and sufficient contribution to the scale. Thus, the item analysis results provide strong support for the test's construct validity and demonstrate its suitability for measuring 9th-grade students' academic achievement in organic matter.

Table 1. Item Analysis Findings.

Item Number	Pj	rjx	Sig.	t	Item-Total Correlation	Explanation
1	0.46	0.81	0.003	-16.606	0.524	
2	0.70	0.43	0.000	-6.214	0.300	
3	0.46	0.73	0.013	-12.564	0.492	
4	0.55	0.33	0.016	-4.110	0.333	
5	0.29	0.50	0.000	-7.723	0.214	
6	**0.47**	**0.34**	**0.157***	**-4.295**	0.357	Eliminated
7	0.43	0.69	0.000	-11.286	0.471	
8	0.34	0.31	0.000	-4.122	0.348	
9	0.56	0.26	0.010	-3.148	0.243	
10	**0.41**	**-0.04***	**0.318***	**.513**	**-0.077***	Eliminated
11	**0.49**	**0.63**	**0.388***	**-9.500**	0.443	Eliminated
12	0.24	0.21	0.000	-3.065	0.218	
13	**0.49**	**0.47**	**0.704***	**-6.280**	0.381	Eliminated
14	0.23	0.37	0.000	-5.793	0.287	
15	0.24	0.37	0.000	-5.645	0.239	
16	0.38	0.67	0.000	-11.266	0.511	
17	0.32	0.44	0.000	-6.326	0.425	
18	0.31	0.29	0.000	-3.799	0.308	
19	0.36	0.64	0.000	-10.543	0.414	
20	**0.16***	**-0.10***	**0.001**	**1.600**	**-0.173***	Eliminated
21	**0.21**	**0.16***	**0.000**	**-2.322**	**0.108***	Eliminated
22	0.32	0.21	0.000	-2.769	0.236	
23	0.40	0.29	0.000	-3.581	0.226	
24	0.28	0.21	0.000	-2.891	0.208	
25	0.33	0.31	0.000	-4.171	0.313	
26	**0.14***	**0.07***	**0.013**	**-1.232**	**-0.100***	Eliminated
27	**0.36**	**-0.13***	**0.004**	**1.583**	**-0.042***	Eliminated
28	0.38	0.30	0.000	-3.820	0.239	
29	**0.29**	**0.07***	**0.067***	**-0.925**	**0.089**	Eliminated
30	0.30	0.43	0.000	-6.214	0.447	
31	0.36	0.39	0.000	-5.139	0.253	
32	**0.46**	**0.23**	**0.051***	**-2.769**	**0.145***	Eliminated
33	0.24	0.30	0.000	-4.438	0.266	
34	0.36	0.63	0.000	-10.208	0.487	
35	**0.12***	**0.07***	**0.009**	**-1.292**	**0.022***	Eliminated
36	0.31	0.29	0.000	-3.799	0.309	
37	0.36	0.64	0.000	-10.543	0.516	
38	**0.24**	**-0.23***	**0.000**	**3.248**	-0.303	Eliminated
39	0.44	0.46	0.003	-6.089	0.375	
40	0.43	0.83	0.000	-17.980	0.542	
41	**0.25**	**0.13***	**0.000**	**-1.764**	**0.014***	Eliminated
42	**0.36**	**0.20**	**0.000**	**-2.507**	**0.134***	Eliminated
43	**0.51**	**0.76**	**0.608***	**-13.619**	0.511	
44	**0.21**	**-0.01***	**0.679***	**0.207**	**-0.143***	Eliminated
45	0.42	0.41	0.000	-5.429	0.332	
46	0.35	0.36	0.000	-4.743	0.343	
47	0.36	0.30	0.000	-3.854	0.342	
48	**0.21**	**0.16***	**0.000**	**-2.322**	**0.088***	Eliminated
49	0.31	0.41	0.000	-5.904	0.402	
50	0.41	0.39	0.000	-5.014	0.424	
51	0.31	0.54	0.000	-8.466	0.444	
52	0.24	0.30	0.000	-4.438	0.313	
53	0.46	0.49	0.022	-6.559	0.385	
54	**0.24**	**-0.13***	**0.000**	**1.800**	**-0.100***	Eliminated
55	**0.06**	**-0.04***	**0.038**	**1.030**	**-0.129***	Eliminated
56	0.35	0.24	0.000	-3.093	0.209	
57	0.30	0.34	0.000	-4.739	0.426	
58	**0.26**	**0.14***	**0.000**	**-1.946**	0.237	Eliminated
59	0.41	0.39	0.000	-5.014	0.326	
60	**0.13***	**0.00***	**1.000***	**0.000**	**-0.053***	Eliminated

** Unwanted values according to item analysis.*

Table 2. Content Framework of the 9th-Grade Organic Matter Topic and the Questions it Covers.

			Number of Questions
Carbohydrates	Monosaccharide	ribose.	Q1. Q8
		Deoxyribose.	Q1. Q8
		fructose.	Q4. Q8
		Glucose.	Q1. Q2. Q8
		Galactose	Q4. Q8
	Disaccharide	sucrose	Q3. Q4. Q8
		Maltose	Q3. Q4. Q8
		Lactose	Q3. Q4. Q8
	Polysaccharides	Glycogen	Q5. Q8
		Starch	Q5. Q8
		Cellulose	Q5. Q7. Q8
		Chitin	Q5. Q6. Q8
Lipids	Fatty Acids		Q12. Q13. Q14
	Triglycerides		Q10. Q14
	Phospholipids		Q11. Q14
	Steroids		Q9. Q14
Proteins	Structure of Amino Acids		Q15. Q16.
	Enzymes	Simple and Compound Enzymes	Q21. Q22. Q17
		Activation Energy	Q20. Q17
		Enzyme-Substrate Relationship	Q19. Q17
	Factors Affecting Enzymatic Reactions		Q18. Q17
Nucleic Acids	Structure of DNA and RNA		Q23. Q.S26. Q27. Q28. Q29
Vitamins	Fat Soluble Vitamins		Q31. Q32. Q34. Q35. Q37. Q38. Q39. Q40
	Water Soluble Vitamins		Q31. Q33. Q35. Q36. Q37. Q38

Results

Item analysis results led to the elimination of 20 items from the original 60-question test due to low difficulty indices, poor discrimination, non-significant upper-lower group differences, and low item-total correlations. The eliminated items are listed in **Table 1** along with the reasons for elimination.

As a result of the analyses, the difficulty index values of the 20th, 26th, 35th, 55th, and 60th items were eliminated from the item pool because they had a value below 0.20 and were considered very difficult, considering the criteria stated above (**Table 1**). The average difficulty index of the test was calculated as 0.33. When this value is considered, it can be said that the test is of medium difficulty.

Item discrimination indexes were calculated for each item in OSAT. As a result of the analysis, it was calculated that the questions 10, 20, 21, 26, 27, 29, 35, 38, 41, 44, 48, 54, 55, 58, and, 60, which were below 0.20 were not discriminatory at the desired level (**Table 2**). The average discriminatory

index of the test was calculated as 0.32. Considering this value, it can be said that the discriminatory power of the test is high.

The results of the independent samples t-test were interpreted as follows: the items with a significance level value less than 0.05 should be included in the test, and since there was no significant difference in those with a significance level value greater than 0.05, these items should be removed from the test. As a result of the analysis, it was observed that the questions with a significance level value greater than 0.05 were questions 6, 10, 11, 13, 29, 32, 43, 44, and 60 (**Table 2**). Based on these results, these questions were removed from the test.

According to the criterion, items 10, 20, 21, 26, 27, 29, 35, 38, 41, 42, 44, 48, 54, 55, and 60 with an item-total correlation value below 0.20 were eliminated.

The KR-20 reliability coefficient was calculated as 0.84 for the 40 questions in the OSAT. This value shows that the reliability of the test is high. According to these results, 20 questions were removed from the test by accepting that they did not serve the purpose. The subject of Organic Matter, which is included in the MoNE (2024) program Turkish Century Education Model, and the content framework of this subject were taken as basis in the development of the test. The prepared test included more than one question covering all subheadings of the subject. Since there was more than one question in the test, removing inappropriate questions did not pose a problem in terms of content validity. The distribution of the questions in the final test according to the content framework is shown in **Table 2**.

The final version of the achievement test is presented in **Appendix 1**.

Discussion and Conclusion

Multiple-choice achievement tests are indispensable tools that are frequently used in both the Turkish education system and international measurement and evaluation practices. The fact that student placement exams for secondary and higher education institutions in Turkey are largely conducted with multiple-choice tests necessitates students to become familiar with this type of test structure and develop relevant skills (Gönen et al., 2011).

The Ministry of National Education (MoNE) makes curriculum changes at certain periods according to the needs of the education process. In this context, the measurement and evaluation tools used in schools should be updated in accordance with the new curriculum (Akgun & Gul, 2024). In addition, in order to prevent the concern of repeating the items when the same tests are used every year, the test contents should be revised for a while and the test should continue to be developed by adding new pilot items to the test (Ansley, 1997).

Item analysis is a basic process that evaluates the contribution of the items in a test to the measurement function (Khairani & Shamsuddin, 2016). In this study, item difficulty index and item discrimination power was calculated for test items. Items with an item difficulty index value below 0.30 were evaluated as "very difficult", between 0.30 and 0.80 as "medium level", and above 0.80 as "very easy"; it was suggested that very easy and very difficult items should be revised on the grounds that they could reduce discrimination. In terms of discrimination power, items between 0.00 and 0.19 were classified as weak, between 0.20 and 0.29 as acceptable but open to improvement, and between 0.30 and 0.39 as having a good level of discrimination (Azzopardi & Azzopardi, 2019).

In addition to the item analysis, the mean difference between lower and upper groups of the test and the correlation values of each item were examined, and it has been suggested in recent test development studies that the difference between the mean scores should be calculated after the lower and upper 27% groups of the sample are created (Demir et al., 2016; Özkan & Eryılmaz Muştu, 2018). Therefore, it can be said that the developed test has the feature of distinguishing between those who know and those who do not.

Initially, 60 questions were prepared, but as a result of the item analysis, some items were eliminated in order to make the test more valid and reliable. As a result of this process, an achievement test consisting of 40 questions that better reflect the scope of the test was created. As a result of the analysis, the achievement test consisting of 40 questions that reflect the scope of the curriculum took its final form. The average item difficulty of the test was calculated as 0.33; and the average item discrimination was calculated as 0.32. These values show that the test is at the desired level in terms of measuring adequacy at the item level (Gömleksiz & Erkan, 2010).

In order to determine the general reliability of the test, the KR-20 (Kuder-Richardson Formula 20) coefficient was calculated with the data obtained from the test applied to a different student group of 204 people. KR-20 is an internal consistency coefficient widely used, especially in tests scored as true-false and is preferred especially when working with items whose difficulty levels are calculated (Bolat & Karamustafaoğlu, 2019). As a result of the analysis, the KR-20 reliability coefficient of the test was found to be 0.84. This value reveals that the test is a highly reliable measurement tool. In the achievement tests developed in the literature, tests with high reliability values are generally encountered, and in this respect, this study also offers high reliability values in line with the existing literature (Baladın Duman, 2019; Çelik & Bayrak, 2025; Ortaakarsu & Sülün, 2025; Şen & Eryılmaz, 2011; Özkılıç et al., 2023). This value indicates a high level of reliability compared to similar tests previously developed in the literature. For example, the Cronbach's alpha coefficient for the "Basic Components of

Living Things" achievement test developed by Şakir (2013) was reported as 0.73. This comparison demonstrates that the test developed in the current study offers higher reliability and a strong measurement adequacy compared to similar studies. Furthermore, as mentioned in the introduction, the need for a multiple-choice measurement tool specifically for the topic of "Organic Matter" that has been extensively tested for validity and reliability is evident in the literature. In this context, this achievement test, whose content validity is supported by expert opinions and whose construct validity is demonstrated through item analysis, fills this gap in the literature and provides a reliable assessment tool for teachers and researchers.

As a result of the validity and reliability analyses conducted during the test development process, the distribution of the achievement test consisting of a total of 40 questions according to the content framework and subheadings was determined. The content of the test includes 8 questions on the topic of "Carbohydrates", 6 on the topic of "Fats", 8 on the topic of "Proteins", 7 on the topic of "Nucleic Acids" and 11 on the topic of "Vitamins". This distribution ensures that the content framework and subheadings in the curriculum are covered comprehensively. It is natural that the questions are not distributed completely equally among the outcomes, as they may vary depending on the importance and scope of each outcome in the teaching process (Kargın and Gül, 2021). In this context, it is thought that the test in its current form comprehensively measures all the contents in the curriculum and that the outcomes are evaluated as necessary. In this context, the developed achievement test can be used in educational practices as a tool that can both measure the content of the curriculum in a comprehensive manner and provide reliable results. In this context, it is thought that the developed achievement test will be useful for researchers, teachers and students. In addition, the following suggestions can be made based on the current research:

- It is recommended that the test be applied to different school types and various student groups, and validity and reliability analyses be conducted on a larger sample.
- Educational curricula and teaching programs may change, so it is recommended that the test be reviewed and updated periodically.
- The scope of the test can be expanded by using different measurement and evaluation techniques.
- By following a similar test development process, valid and reliable achievement tests appropriate for the achievements in other subjects can be developed.

The improvements made in line with these suggestions will ensure that the achievement tests developed will be more comprehensive, valid, reliable, and will contribute to more effective measurement processes in education.

Limitations

This study has several limitations that must be acknowledged. First, the study sample was limited to 9th-grade students in public high schools in a specific region of Türkiye, which may limit the generalizability of the results to broader student populations. Second, although expert review and item analysis were conducted meticulously, potential biases in student item interpretation may still exist. Additionally, the use of a paper-based, classroom-administered test may have influenced student performance due to environmental or motivational factors. Therefore, these limitations should be considered when administering the test in different educational contexts.

The final version of the achievement test is presented in **Appendix 1**.

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Appendix 1

1. Which of the following statements about ribose, deoxyribose, and glucose is incorrect?

A) They are 6C carbohydrates.
B) Ribose participates in the structure of RNA and ATP.
C) Deoxyribose means ribose with one oxygen atom removed.
D) Unlike glucose, ribose, and deoxyribose, it is used as an energy source.
E) Deoxyribose participates in the structure of DNA.

2. What is the common function of glucose in all living things?
A) It participates in the structure of the cell wall.
B) It participates in the structure of organelle membranes.
C) It is used in aerobic respiration.
D) It synthesizes ATP from the chemical bond energy in its structure.
E) It participates in the structure of the nuclear membrane.

3. Which of the following is false about disaccharides?
A) Every disaccharide is water-soluble.
B) Every disaccharide contains a glycosidic bond.
C) Maltose and sucrose are types of plant disaccharides.
D) Lactose is an animal disaccharide.
E) Every bacterium synthesizes all types of disaccharides.

4. Which of the following statements is incorrect?
A) Galactose ——— Glucose + Fructose + H_2O
B) Maltose ——— Glucose + Glucose + H_2O
C) Lactose ——— Glucose + Galactose + H_2O
D) Sucrose ——— Glucose + Fructose + H_2O
E) A disaccharide is a molecule composed of two sugar units.

5.Which of the following statements regarding polysaccharide types is incorrect?
A) Human cells do not produce the enzyme that hydrolyzes starch.
B) Beneficial bacteria living in the rumen of ruminant herbivorous mammals aid in cellulose digestion.
C) The glycogen molecule can be produced in bacterial, fungal, and animal cells.
D) Chitin is found in the exoskeletons of arthropods and the cell walls of fungi.
E) The difference between polysaccharide types stems from the different positions of the glycosidic bonds.

6. Chitin, which is part of the exoskeleton of arthropods such as insects and spiders, contains which of the following elements, unlike other types of polysaccharides?
A) Nitrogen
B) Oxygen
C) Hydrogen
D) Carbon
E) Phosphorus

7. The most stable structural form of some polymers like cellulose is to form long, straight chains. When several chains lie side by side, the network stabilized by hydrogen bonds between and within the chains creates a regular, stationary, and strong fiber.

Accordingly, this structure of cellulose:
I. Has made it an important material of civilization in this century.
II. Has enabled its use in the production of products such as paper and cardboard.
III. Has provided advantages in the production of products such as insulation materials and many other construction materials.
IV. Has facilitated the production of industrial products with high water content.

Which of the above statements are valid?

A) Only I
B) Only II
C) Only III
D) I, II, and III
E) I, II, III, and IV

8. Which of the following is false about carbohydrates?
A) Some types participate in the structure of nucleic acids.
B) They can form compound molecules with fats and proteins.
C) All types can be used as respiratory raw materials.
D) Glycosidic bonds are formed between their monomers during polymerization.
E) They participate in the structure of all living cells.

9. Which of the following statements about steroids is incorrect?

A) They increase membrane permeability by incorporating them into the cell membrane.
B) They are used in the activation of provitamin D in the human body.
C) They contribute to the structure of the myelin sheath, which increases the speed of conduction in nerve cells.
D) They act as a regulator by participating in the structure of sex hormones.

E) They form the skeleton of the cell membrane by lining it up in a double layer.

10. Which of the following statements about triglycerides is false?
A) They provide more energy than the same amount of carbohydrates and proteins.
B) They contain three ester bonds.
C) They contain three glycerol molecules.
D) Whether they are saturated or unsaturated depends on the properties of the fatty acids they contain.
E) Some of the fatty acids included in their structure are synthesized in the human body, while others are obtained through the diet.

11. Regarding phospholipids:

I. They are the main components of the plasma membranes of prokaryotic and eukaryotic cells.
II. Some are water-soluble while others are not.
III. The membrane allows water-soluble molecules to pass but not fat-soluble ones.

Which of the following statements are correct?

A) Only I
B) Only II
C) Only III
D) I and II
E) I, II, and III

12. A fat molecule is generally formed by combining which of the following substances?

A) One molecule of glycerin and three molecules of fatty acids
B) One molecule of glycerin and two molecules of fatty acids
C) Two molecules of glycerin and two molecules of fatty acids
D) Two molecules of glycerin and one molecule of fatty acids
E) Two molecules of glycerin and four molecules of fatty acids

13. Cooking oils, which are liquid at room temperature, can be converted into solid fats by chemical means. Which of the following has changed during this process?

A) The structure of glycerol

B) The number of ester bonds

C) The number of C atoms in fatty acids

D) The number of O atoms in fatty acids
E) The number of H atoms in fatty acids

14. Fats in the human body are used for which of the following functions?
I. Constructive–repair
II. Storage
III. Energy supply
IV. Heat insulation
Which of these functions apply?
A) I, II, and III
B) I, II, and IV
C) I, III, and IV
D) II, III, and IV
E) I, II, III, and IV

15. Although proteins are made up of 20 different amino acids, thousands of different proteins are synthesized in living organisms. Regarding this, which of the following statements are correct?

I. The repeated use of the same types of amino acids
II. Different sequences of amino acid types
III. Amino acids are connected by peptide bonds
IV. Water is released during protein synthesis

Which of these statements are true?

A) Only I
B) Only II
C) I and II
D) I, II, and IV
E) I, II, III, and IV

16: Which of the following statements about the structure of amino acids is incorrect?

A) The carboxyl group gives the molecule its acidic properties.
B) The carboxyl group and the amino group combine to form a polypeptide.
C) The type of amino acid is determined by the radical group.
D) All amino acid types can be produced by plants.
E) A peptide bond exists between the central carbon atom and the radical group.

17. Which of the following is not one of the basic functions of proteins in living things?
A) Structural and mechanical functions
B) Receptor function
C) Immune function
D) Enzyme and hormone function

E) Hereditary function

18. The sweet taste of freshly harvested corn is due to the high sugar levels in the corn kernels. Corn purchased from the grocery store (even days after harvest) is not sweet. To preserve the sweetness of fresh corn, the hulled corn cob should be immersed in boiling water for a few minutes ("withering") and then cooled in cold water. Corn that has undergone a specific treatment and been stored in the refrigerator by this method retains its sweetness.

Therefore, which of the following is the reason why corn subjected to the withering process retains its sweetness?

A) The enzyme that converts sugar to starch is denatured by heat.
B) Enzymes operate within specific pH ranges.
C) Enzymes initiate reactions.
D) Enzymes are inactive at high temperatures.
E) Enzymes are active in a certain amount of aqueous medium.

19. After an enzyme reacts with a substrate and dissociates, which of the following can it perform?
I. Reacting with another substrate of the same type
II. Forming a stable compound with intermediate products formed during the reaction
III. Reacting with the products in the environment to regenerate the original substrate
Which of the statements are correct?
A) Only I
B) Only III
C) I and III
D) II and III
E) I, II, and III

20. Which of the following best describes activation energy?

A) It is the minimum amount of energy required for any chemical reaction to begin.
B) It is the minimum amount of energy required for enzymes to detach from their substrate.
C) It is the maximum amount of energy supplied by the enzyme to initiate the reaction.
D) It is the amount of energy generated by light energy in all living things and consumed in the production of organic food.
E) It is the amount of energy remaining in the structure of the products after an enzymatic reaction has completed.

21. Regarding conjugated enzymes:
I. The protein part is called an apoenzyme.
II. If the auxiliary part is an organic substance, it is called a cofactor.
III. The apoenzyme is inactive and cannot function without the auxiliary part.

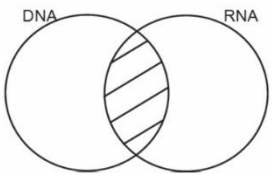
Which of the following statements is incorrect?

A) I only
B) II only
C) III only
D) I and III
E) I, II, and III

22. Which of the following is a type of enzyme that has only a protein structure and does not contain an auxiliary group (coenzyme or cofactor)?

A) Holoenzyme
B) Apoenzyme
C) Simple enzyme
D) Coenzyme
E) Cofactor

23.



Which of the following can be written in the shaded area?
A) Being double-chained
B) Containing deoxyribose sugar
C) Being self-replicating
D) Being involved in protein synthesis
E) Carrying uracil base

24. Which of the following is incorrect regarding the bases found in the structures of DNA and RNA?
A) Double-ringed bases are called purine bases.
B) Single-ringed bases are called pyrimidine bases.
C) Hydrogen bonds are formed between the double-ringed bases in DNA.
D) The double-ringed bases in RNA are adenine and guanine.
E) Purine bases are found in both DNA and RNA

25. Which of the following events occurs last during DNA replication?

A) The separation of the two strands of DNA that will replicate themselves

B) The bonding of nucleotides with hydrogen bonds and their three-dimensional structure
C) The attachment of pentose sugars to nitrogenous organic bases
D) The formation of a new DNA strand
E) The attachment of phosphates to nucleosides

26. During the replication of DNA, which of the following occur?
I. An increase in the number of hydrogen bonds
II. A decrease in the number of free nucleotides
III. Release of energy
IV. An increase in the number of phosphodiester bonds
Which of the statements above is/are correct?
A) I only
B) I and II
C) I, II, and IV
D) I, III, and IV
E) II, III, and IV

27. Which of the following is a common feature of all RNA types?
A) Participation in the structure of the ribosome
B) Containment of hydrogen bonds
C) Transport of amino acids to the ribosome
D) Transport of genetic information in DNA to the ribosome
E) Synthesis via DNA

28. Which of the following is not one of the structures found together in ATP and RNA?

A) Adenine
B) Ribose
C) Ester bond
D) Phosphoric acid
E) Guanine

29. Which of the following is the Purine/Pyrimidine ratio in a DNA molecule when the number of hydrogen bonds and the total number of Guanine nucleotides are known?

A)1
B)1/2
C)) 1/3
D) 1/4
E) 3/4

30. Vitamins are divided into two classes: water-soluble and fat-soluble.

Accordingly, which of the following statements is true about water-soluble vitamins?

I. They can be synthesized entirely by plants.
II. Excess amounts are generally excreted in the urine.

III. They serve as coenzymes in enzymatic reactions
.A) Only I
B) Only II
C) Only III
D) II, III
E) I, II, III

31: Which of the following statements about vitamins is incorrect?
A) They can all be absorbed without hydrolysis.
B) Vitamin E is stored primarily in fat tissue.
C) Excess B vitamins, except vitamin B12, can be excreted in the urine.
D) Vitamin K can also be synthesized by intestinal bacteria.
E) Fat-soluble vitamins provide more energy

32.

• It is the first vitamin group to show a deficiency in the body.
• It is one of the vitamins synthesized by beneficial bacteria living in the human large intestine.
• A deficiency can cause infertility.
• It is a vitamin that plays a role in cell renewal, resistance to microorganisms, and protection from sunlight.

Accordingly, which of the following vitamins is not mentioned?
A) B B) A C) D D) E E) C

33. Regarding water-soluble vitamins, which of the following statements are correct?

I. They cannot pass through the cell membrane.
II. They are used as an energy source only in urgent situations.
III. They are not stored in the body.

A) I only
B) III only
C) I and II
D) II and III
E) I and III

34. Which of the following statements is definitely true for a vitamin that can be stored in liver cells when consumed in excess of the required amount?

A) It must be transported via special carrier proteins in the cell membrane.
B) It is taken in inactive form and activated by special enzymes within the body.

C) If it is insufficient, blindness occurs due to the malfunctioning of the photoreceptors in the eye.

D) It polymerizes by forming covalent bonds with various organic substances.
E) It does not form any of the urine components in healthy people.

35. Vitamin-rich foods should be consumed fresh. Factors such as heat, light, storage, and contact with metals can damage the structure of vitamins. Therefore, vitamins can be compared to which of the following food groups?

A) Proteins B) Carbohydrates D) Minerals C) Fats E) Salts

36. Which of the following is a water-soluble vitamin?

A) Vitamin A B) Vitamin E D) Vitamin D C) Vitamin C E) Vitamin K

37. Which of the following is false about vitamins?

A) They are generally sourced from plants.
B) They are used as energy sources.
C) They pass into the bloodstream without being digested.
D) They are not incorporated into cell structure.
E) They form the coenzyme portion of compound enzymes.

38. If the following types of vitamins are deficient, which of the following options correctly matches them with the associated diseases?

I. Vitamin B
II. Vitamin C
III. Vitamin K
IV. Vitamin A

Associated diseases:
a. Anemia
b. Scurvy
c. Blood clotting disorders
d. Night blindness

	I	II	III	IV
A	**a**	**b**	**c**	**d**
B	**a**	**c**	**b**	**d**
C	**b**	**d**	**a**	**c**
D	**b**	**c**	**d**	**a**
E	**c**	**a**	**d**	**b**

39. Which vitamin deficiency can cause night blindness?
A) Vitamin E
B) Vitamin K
C) Vitamin B1
D) Vitamin A
E) Vitamin C

40. Which vitamin strengthens the immune system and acts as an antioxidant?
A) Vitamin D
B) Vitamin B12
C) Vitamin E
D) Vitamin K
E) Vitamin B1

ANSWER KEY
1 : A
2: D
3: E
4: A
5: E
6: A
7: D
8: C
9: A
10: C
11: D
12: A
13: E
14: E
15: C
16: E
17: E
18 :A
19: A
20: A
21: B
22: C
23: D
24: C
25: B
26:C
27: E
28: E
29: A
30: D
31: E
32: C
33: B
34: C
35: A
36: C
37: B
38: A
39. D
40. C