C1 We burn sugar – Cellular respiration and respiratory chain

This is a typical introductory experiment to the overall topic of energy supply in the human body, cellular respiration, and the respiratory chain. Three straightforward subexperiments (combustion of sugar with and without a catalyst and verification of water and carbon dioxide [CO_2] in exhaled air) suggest the obvious explanation that sugar is also burned in the human body. The analogy between heterogeneous catalysis in the combustion of sugar in the experiment and biocatalytic combustion in the body leads on to the topic of energy metabolism and cellular respiration in the human body. In the age range up to 16 years, a qualitative interpretation of the experimental results may suffice. In specialized teaching in the age group 16+, teachers must go into the subject matter in considerably more detail independently of the experiment, provided adequate prior knowledge can be assumed. The materials and apparatus supplied allow eight groups of students to conduct the experiments simultaneously.

1 Main question

Sugar (sucrose) is a carbohydrate and as such burns to produce water and carbon dioxide. But how does sugar "burn" in the human body and is it really the main provider of energy in human metabolism? From a subexperiment on catalytic sugar combustion through two additional subexperiments on detection reactions for the products of cellular respiration, we arrive at the topic of the energy transfer at the cellular level. Students are given insight into the oxidative processes of degradation in the human body at the cellular level that convert the energy of high-energy nutrients into ATP. The constituents of food, especially carbohydrates and fats, are decomposed for this purpose via the three reaction paths of cellular respiration – glycolysis or β-oxidation, citric acid cycle, and oxidative phosphorylation. The low-energy products water and carbon dioxide occur due to reaction with oxygen. The detection reactions show these two degradation products that are dissipated in the ambient air during exhalation.
With respect to the overall topic "Digestion and energy balance of humans" there are two additional experiments: C2 (Carbohydrates as providers of energy for metabolism – Starch and sugar) and C3 (How does human digestion break down fats? – Saponification of edible oil).
Regarding practical methodology, students will be familiarized with the systematic variation of variables when experimenting in order to be able to trace the detection back to precisely one substance.

2 Integrating the experiment into the teaching context

2.1 Basic principles

These subexperiments relating to the topic of health open up an experimental avenue to the overall topic of nutrition, respiration, substance transport, and energy conversion. Special attention should be paid during teaching to establishing correlations between the various subject areas. What do respiration and blood circulation have to do with nutrition? The relationship between absorption, transport, and release of substances and energy should be dealt with explicitly.
The oxidative processes in the cell are difficult for younger students in particular to imagine since they are not visible and can be detected only indirectly through the development of body heat, for example, during physical activity. Children also have little access to substance conversion at the particle level; as they imagine it, atoms are ground down by the teeth, digested in the intestines, or "destroyed" in the lungs. Understanding metabolic processes in the cell, especially the respiratory

chain at the molecular level, requires a knowledge of reaction equations and combustion processes as a minimum for qualitative treatment. Knowledge of redox reactions is required for dealing with the subject in more depth.
Basic knowledge of acids and bases is a prerequisite for understanding the acid reaction of carbon dioxide in water.

2.1.1 Qualitative access at age levels 10 to 16

Cane sugar (sucrose) burns with oxygen to produce water and carbon dioxide. We can verify this theoretically via a reaction equation.

$$C_{12}H_{22}O_{11} + 12\ O_2 \rightarrow 11\ H_2O + 12\ CO_2$$

However, we see in subexperiment 1 that in practice it is not that easy to burn sugar. You need a catalyst. It's a similar case in the human body, where the chief energy-providing sugar, glucose (which is partly a product of sucrose conversion), releases its energy only via a complex chain of chemical reactions. These are catalyzed by enzymes (enzyme = biocatalyst).
In our second subexperiment we verify this fact by way of cellular respiration. Cellular respiration combines the processes of nutrition and digestion with gas exchange. Transport of the basic substances for cellular respiration is assured by the blood. This brings the high-energy compounds (for example, carbohydrates, glucose, fats) that occur as a result of the mechanical and enzymatic breakdown of food in the mouth, stomach, and intestine as well as oxygen to the tissues in the body. The high-energy compounds and the oxygen pass from the blood into the tissue cells, where they react in a complex sequence of reactions to produce the low-energy compounds water and carbon dioxide. The high-energy compounds are therefore oxidized ("burned") by the oxygen. The reaction energy is used to build up an electrochemical gradient and subsequently to synthesize ATP.
Alternatively, fatty acids can also be oxidized by oxygen.
The following reaction equation can be formulated for the oxidation of high-energy compounds using the example of glucose:

$$\text{Glucose} + \text{Oxygen} \rightarrow \text{Carbon dioxide} + \text{Water} + [\text{Energy}]$$

$$C_6H_{12}O_6 + 6\ O_2 \rightarrow 6\ CO_2 + 6\ H_2O + [\text{ATP} + \text{heat}]$$

2.1.2 Detailed explanation of the processes for the age group 16+

The metabolic processes for oxidation of the high-energy compounds take place in different parts of the cell:

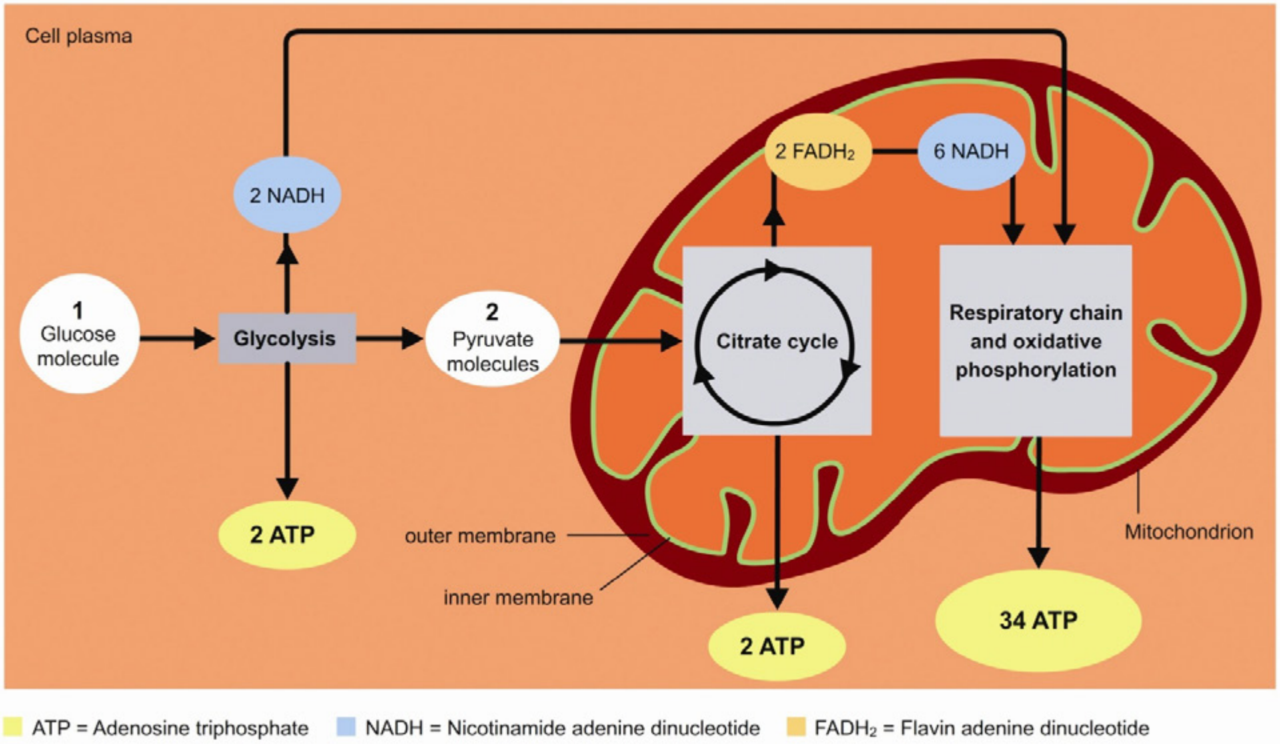

Fig. 1: Overview of cellular respiration.

In **glycolysis**, the carbon skeleton of a glucose molecule with six carbon atoms is transformed into two molecules with three carbon atoms; the resulting compound is called pyruvate. Glycolysis occurs in the cell cytoplasm.

The pyruvate is transported into the mitochondria. In preparation for the **citrate cycle**, the carbon skeleton of the pyruvate is reduced from three to two carbon atoms and bonded to the coenzyme A. This reaction produces acetyl-coenzyme A (or acetyl-CoA or activated acetic acid). In the process, carbon dioxide and hydrogen are released. The hydrogen is transferred to the coenzyme NAD^+, resulting in $NADH+H^+$. In the citrate cycle, the remaining carbon skeleton with two carbon atoms on the acetyl coenzyme A is oxidized. Hydrogen and carbon dioxide are again released, and the hydrogen is once again transferred to the coenzyme NAD^+. $NADH+H^+$ is oxidized on the inner mitochondrial membrane with the inhaled oxygen in the air:

$$2\ H_2 + O_2 \rightarrow 2H_2O + \text{Energy}$$

The reaction corresponds formally to the oxyhydrogen reaction and releases large amounts of energy. To prevent damage to cells, this redox reaction occurs via the **respiratory chain**. Electrons are transferred between the complexes via several successive protein complexes in the inner mitochondrial membrane. In this way, only a part of the total reaction energy is released at a time. The final protein complex in the respiratory chain transfers the electrons together with protons (H^+ ions) from the mitochondrial matrix to oxygen, whereby water is generated as the end product of the respiratory chain.

The reaction energy is used at the protein complexes of the respiratory chain to get protons from inside the mitochondrion into the gap between the two mitochondrial membranes. This creates a proton gradient across the inner mitochondrial membrane that acts as a temporary energy store. If protons flow back along their gradient from the gap between the two mitochondrial membranes into the mitochondrial matrix through enzyme ATP synthesis, the released energy is used for forming ATP.
To sum up: The reaction products carbon dioxide and water occur in two different metabolic processes. While carbon dioxide occurs already in the citrate cycle, water is formed at the end of the respiratory chain. The reactions in the mitochondria also produce the reduced coenzyme $NADH+H^+$, which oxidizes to provide the energy for ATP synthesis.

2.2 Relevance to the curriculum

Whereas the anatomy and physiological processes at the organ level involved in eating, digestion, and respiration are covered by the curriculum in the age group from 12 to 16, the metabolic physiological processes at the cell level are not usually dealt with until the age group 16+, drawing on prior knowledge of general and organic chemistry.
Nevertheless, the detection reactions for the degradation products of metabolism can be used as qualitative experiments starting from the age group 10+. In this case, it is advisable to have students brush up on prior knowledge with respect to simple combustion processes (candles, burning of sugar with or without a catalyst).
Combustion processes can be viewed from the chemical perspective. Oxidation and reduction should be considered as electron transfers in order to be able to understand their application at the cellular respiration level. The interdisciplinary component is thus provided through oxidation and reduction within the biological context of cellular respiration.

Topics and terms: acid-base reaction, ATP, biocatalyst, catalysis, carbohydrates, carbon dioxide, cellular respiration, citrate cycle, citric acid cycle, combustion process, cytoplasm, energy provider, enzyme, fat, glycolysis (β-oxidation), metabolism, mitochondria, NAD, oxidative phosphorylation, oxygen, oxyhydrogen reaction, pH value, protons, pyruvate, redox reaction, respiratory chain, substance transport, sugar, water

2.3 Skills

The students will ...

- be able to apply the principle of sugar combustion to the metabolic processes.
- understand the cell as a system in which different metabolic processes interact.
- be able to summarize the basic principles of energy conversion through catabolism.
- be able to roughly explain cellular respiration and formulate the gross equation.
- be able to explain the connection between gas exchange in the lungs and cellular respiration.
- be able to meaningfully plan, implement, and evaluate suitable qualitative experiments to verify products of metabolism.

2.4 Explaining the experiment in the teaching context

2.4.1 Subexperiment 1: Sugar can be burned

The purpose of this subexperiment is to brush up students' prior knowledge of combustion processes. It also illustrates the particular feature of sugar combustion, namely that sugar cannot be ignited and does not keep on burning. Students will first try unsuccessfully to light a sugar cube. They will then light a sugar cube with paper ash sprinkled over it in a tea light holder, holding a test tube over the flame for a few seconds. We observe that a catalyst is required to burn sugar. During combustion it is apparent that water vapor occurs and condenses on the cold glass surface of the test tube.

Note on igniting the sugar: The igniting of sugar has been successfully tested with the ash from a wide variety of paper types. Nevertheless, occasionally the sugar may fail to ignite if the experimentation instructions are not followed carefully. Encourage your students to work precisely. (Perhaps you should try out the experiment for yourself before you have the students conduct it.) Matches are not suitable for igniting the sugar. The experiment will be successful only if the hot, targeted flame of a gas igniter is used. **Tip:** If the experiment with paper ash does not work, it always works with cigarette ash.

Technical explanation: There are many incorrect explanations for the effect of the ash, such as a "wicking effect." However, since it is not the liquid sugar that burns, but rather the sugar's decomposition gases, this explanation is certainly wrong. The correct explanation is the ash's catalyst effect. On the Web and in literature, there is also the explanation that the iron or iron oxide contained in cigarette ash acts as a catalyst. However, experiments with iron and iron oxide did not confirm this. Another suggestion is to use manganese dioxide (MnO_2) instead of ash. However, manganese dioxide probably acts more as an oxygen-containing oxidizing agent and not as a catalyst. So what does the ash from paper or cigarettes contain that is effective? Certainly not manganese dioxide. Using activated carbon instead of ash is also questionable from a didactic standpoint. The activated carbon may indeed initially act as a heterogeneous catalyst: The oxygen in the air adsorbs onto the activated carbon, which transfers the oxygen directly to the sugar molecules. However, this process continues only until the activated carbon itself has burned up. The most probable explanation is likely as follows: The alkali oxides contained in many types of ash act as homogeneous catalysts for the hydrolytic decomposition of the sugar at higher temperatures. The low-molecular fragments then form highly combustible gases, which mix with the oxygen in the air and enable a flame that continues to burn autonomously. Because tobacco ash contains more of these alkali oxides than paper ash, it works particularly well.

2.4.2 Subexperiment 2: Verification of reaction products in the air that is breathed: Substance A (water)

The steamed up test tube is an indication of the condensed water vapor in exhaled breath. This can be related directly to the observations from subexperiment 1.

2.4.3 Subexperiment 3: Verification of reaction products in the air that is breathed: Substance B (carbon dioxide)

In this subexperiment, the carbon dioxide content of the solution is systematically varied in the three experimental approaches.

While on the one hand carbon dioxide physically dissolves in water, it also reacts with the water in an acid-base reaction to form hydrogen carbonate (HCO_3^-) and carbonate (CO_3^{2-}) and oxonium ions (H_3O^+):

a) $CO_{2(g)} + 2\ H_2O_{(l)} \rightarrow HCO_3^-{}_{(aq)} + H_3O^+{}_{(aq)}$

b) $HCO_3^-{}_{(aq)} + H_2O_{(l)} \rightarrow CO_3^{2-}{}_{(aq)} + H_3O^+{}_{(aq)}$

Reaction equation 1: Acid-base equilibrium of carbon dioxide, forward reaction.

The oxonium ions formed can be detected by measuring the pH value with pH paper or other indicator – the solution is mildly acidic, with the equilibrium on the side of the starting materials. Carbon dioxide is added, for example, to mineral water. There are essentially two reasons for this: The water remains fresh longer due to the slightly preserving action and the perceived thirst quenching effect is greater. The gas can alternatively also be produced by means of sodium (sodium hydrogen carbonate, $NaHCO_3$) and acetic acid (CH_3-COOH). In this reaction, the reverse reaction of reaction 2.4.3 a) occurs so that carbon dioxide escapes from the solution:

$$HCO_3^-{}_{(aq)} + H_3O^+{}_{(aq)} \rightarrow CO_{2(g)} + 2\ H_2O_{(l)}$$

Reaction equation 2: Acid-base equilibrium of carbon dioxide, reverse reaction of 2.4.3 a).

In cellular respiration, carbon dioxide likewise occurs, which is given off to the ambient air during exhalation. If the exhaled air is introduced into water, the reaction stated in reaction equation 2.4.3 a) also occurs. This solution is therefore also mildly acidic. In this experiment, the reference for the zero point is the sample with pure water.

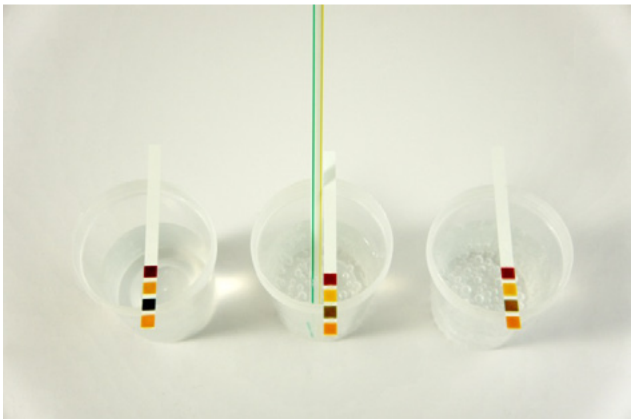

Fig. 2: If the experiment is conducted properly, a reduction in the pH value will be noticeable from the change in color of the green color field in the case of the samples with carbon dioxide (in the center, from exhaled air; on the right, mineral water) compared with pure water (on the left).

The acidic reaction of carbon dioxide in an aqueous solution also serves as an analogy for the transport of carbon dioxide in the blood as a simplified way for teaching about this. About 10% of the carbon dioxide occurring in cellular respiration is physically dissolved in the blood plasma. The vast majority of the hydrated carbon dioxide diffuses into the red blood cells, where it reacts through enzyme catalysis as in reaction equation 2.4.3 a) to produce hydrogen carbonate and protons. Some of the hydrogen carbonate is released from the red blood cells into the blood plasma and transported to the lungs in dissolved form. The protons are bonded to different blood proteins so that the pH value of the blood remains constant to a very large extent (buffer effect of blood). Carbon dioxide from all forms of transport is expelled again from the lungs.

2.5 Experimental variations

The experiments on combustion and on the products of metabolism do not require much material or time and can be integrated into lessons as demonstration or student experiments, which can be conducted individually or in pairs. Because of the complexity of the subject matter, there are teaching methods available to spur students to action. These methods provide additional materials for background information in addition to the experimental approach. Typical examples of this approach could be learning at workstations or group puzzles. These methods are likewise well suited to allow for the different pace of learning and learning progress of individual students.

3 Additional information on the experiment

You will find additional media for preparing or for further study of this experiment on the Siemens Stiftung Media Portal:
https://medienportal.siemens-stiftung.org

Ideas for content:
Organizational overview for the general topic of nutrition, respiration, substance transport, and energy conversion with a list of experiments in the form of a mind map (can also be a work assignment for students at the beginning of the teaching unit)

Ideas for teaching method:
Additional materials for working at a station, e.g., learning dominos, information sheets, and inquiry tasks, models, simulation of the respiratory chain, and ATP synthesis with learning assignments

4 Notes on conducting the subexperiments

4.1 Facilities

The experiments can be conducted by the students on their own under the supervision of a teacher in any well-ventilated classroom.

4.2 Time required

	Preparation	Execution	Analysis and discussion
Subexperiment 1	3 min.	3 – 5 min.	approx. 15 min.
Subexperiment 2	2 min.	2 min.	approx. 15 min.
Subexperiment 3	5 min.	10 min.	approx. 15 min.

4.3 Safety aspects

The students may conduct the experiments only in the presence and under the supervision of the teacher.
The teacher is to point out to the students that the provided materials may be used only according to the respective instructions.
For these experiments, watch out for the following potential dangers and make your students aware of them:

- Working with a flame can result in burns or fires. Before a lighter is used for the first time, the teacher must check that it is working properly and, above all, adjust the flame height.
- Do not allow students to play with fire. In subexperiments 1 and 3, the aluminum bowl can be used as a fireproof base.
- None of the foods provided for the experiments are suitable for consumption.

4.4 Apparatus and materials

Required materials that are not supplied:

- Tap water (a pH value of about 7.0 – 7.5 is perfectly suitable)
- Distilled water, if available
- Mineral water
- Paper
- For subexperiment 1: One lighter per student group (if possible, a gas igniter). Absolutely do not use matches instead of a lighter, because the experiment will not work otherwise.

Supplied:

The apparatus and materials supplied are sufficient to allow **eight** groups of students to conduct the experiments simultaneously.
Safety-relevant materials and apparatus must be tested for proper functioning before being handed out to the students.
The following materials included in the kit are needed for **one** group of students:

Material	Quantity
Bowl, aluminum	1x
Drinking straw	1x
pH test strip, package	1x for entire class
Plant clip (as test tube holder)	1x
Plastic cup, 100 ml	3x
Sugar cube, package	1 cube per group
Tea light or metal cup from a tea light	1x
Test tube, glass, 13 cm	1x

Fig. 3: Apparatus and materials supplied for one group of students.

4.5 Cleanup, disposal and recycling

All apparatus and nearly all materials from the kit can be reused. Therefore, after the students have completed the respective experiment, they should put the apparatus and materials back in the appropriate boxes and return them to the kit. This practice will ensure that you and your colleagues will find everything again quickly the next time the kit is used.
Apparatus that become dirty during the experiment, such as cups, bowls, spoons, and test tubes, should be cleaned before being returned to the kit. We recommend that you have the students do this immediately after they have completed the experiment.
Also make sure that the apparatus are in working order for the next time. For example, recharge used accumulators immediately. (It makes sense to charge the accumulators even if they will not be used for an extended period.)
Materials that cannot be reused, such as used pH test strips and filter paper, should be disposed of properly.
The waste that accumulates during this experiment can be disposed of in the regular trash or poured down the sink.