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# DRINK AND THINK



-  energy drinks, isotonic drinks, caffeine, sugar, effort
  -  chemistry, biology, physics, mathematics
  -  part 3.1: 14–18 years and part 3.2: 8–18 years
- Studies of the components of energy drinks and the dangers they pose to health are appropriate for all ages 8–18 years.

## 1 | SUMMARY

On the market we can see a number of energy, energetic or energising drinks containing recognisable components that can improve the drinker's performance but also present dangers for his or her health. Here we give suggestions on how to teach about these drinks and about methods for finding out their contents and their effects on brain and muscle activity.

## 2 | CONCEPTUAL INTRODUCTION

This teaching unit deals with drinks related to football and sports in general. Today, more and more drinks are being launched on the market in order to improve consumers' physical and mental performance.

These are the main questions of this project:

- What are these drinks made of? How can we analyse their contents?
- What are their effects on mental and physical activity? How can we measure these effects?

This project focuses on three different kinds of drinks:

- Energy drinks: increase your heart rate and blood pressure
- Isotonic drinks: provide sugars and minerals to enhance muscle and brain activity
- Vital drinks: just water

## 3 | WHAT THE STUDENTS DO

### 3 | 1 Energy drinks

Energy drinks are drinks designed to give their consumers an energy boost, using a mixture of different stimulating ingredients. These ingredients include caffeine, an alkaloid that acts as a stimulant and a psychotropic. They may also include taurine, an amino acid whose effects on the human body are still unknown today.

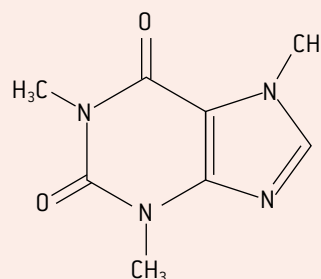
### Biology

First, students of any age can discuss energy drinks and investigate their caffeine content by looking at the labels of some commercial products (for this, the students can just take photos at local stores; they do not even need to buy the drinks). They can do research on the caffeine content and then compare their results with the caffeine content of an espresso and discuss the related health issues.

### Conclusion

Caffeine, whose effects on the human body are well known today, is the ingredient that has by far the greatest effect, whether good or bad, in drinks of this kind.

FIG. 1 Caffeine



A can of an energy drink (250 mL) contains about 80 mg of caffeine, which is approximately the same as a cup of strong black coffee. This amount is very close to the dose where side effects can be expected (100 to 160 mg) and is also very close to the upper limit of daily permissible consumption (for adults 200 mg/day). The risk for athletes lies not in testing positive in an anti-doping control, but in absorbing a toxic dose.

### Chemistry for 14- to 18-year-olds

The analysis of popular commercial products in chemistry laboratory courses is an established method of promoting student engagement, interest and understanding. Many analyses can be done at different levels and using different methods and materials.

### 3 | 1 | 1 Extraction and identification of caffeine

A qualitative analysis using classical thin-layer chromatography can be carried out to verify that energy drinks contain caffeine. First the students will have to extract the caffeine by using an innocuous solvent such as ethyl acetate after a basic treatment in order to solubilise the acids and eventually the tannin.

### Extraction method:

- Take 50 mL of the drink and stir it with a glass rod to degas it if necessary.
- Add a 1 mol/L solution of washing soda (sodium carbonate) while shaking the container, so as to obtain a pH close to 9.
- Extract using 15 mL of solvent and a separating funnel.
- Collect the phase containing the caffeine in a beaker.
- Repeat the extraction with 15 mL of solvent.
- Collect the organic phases and dry them using anhydrous magnesium sulphate.

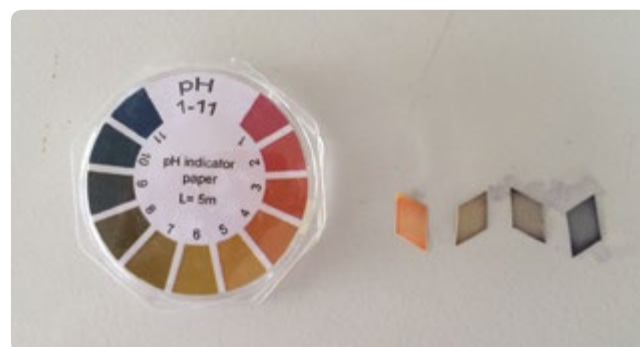


FIG. 2 Checking the basification with pH paper



FIG. 3 Solvent extraction of the caffeine

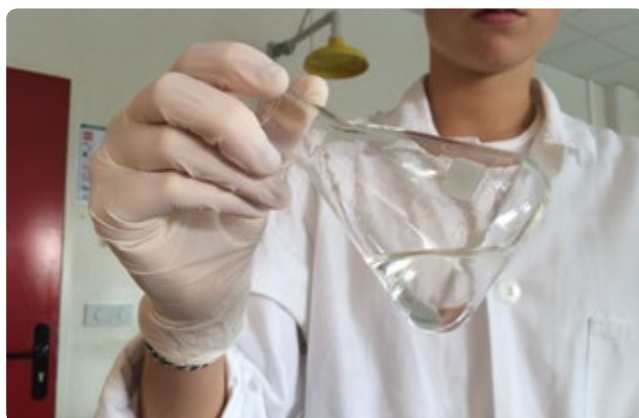


FIG. 4 Drying the organic phase using a desiccant



FIG. 5 Chromatography of the organic phase

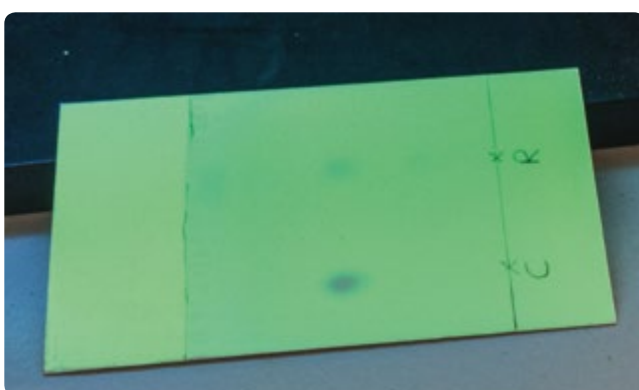


FIG. 6 Visualisation of the chemical species with ultraviolet light

The results of the chromatography must be recorded at the end of this step before the vaporisation of the solvent.

- Eluent (mobile phase) for caffeine: a mixture of formic acid and butyl acetate (30 mL/50 mL)
- Stationary phase: a thin layer of silica
- Visualisation: UV
- Caffeine as a reference dissolved in ethanol or the eluent.

Using chromatography, the students can identify caffeine and another compound that produces a separate spot (indicating that this second compound cannot be neglected in the organic phase after the extraction). After reading the composition of the drinks, students could deduce that this second compound could be a vitamin that has lots of double bonds, particularly B3 or B6.

#### FIG. 7 B6 (pyridoxine) & B3 (niacin or niacinamide)

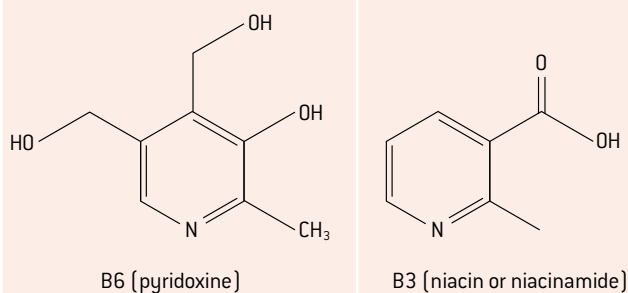


FIG. 8 Evaporation of the solvent using a rotary evaporator (left) · Powder on the side of the flask after the evaporation of the solvent

#### 3 | 1 | 2 Dosage of caffeine

First, an analysis can be carried out using the Beer-Lambert Law.

- The students can determine the spectrum of an aqueous caffeine solution and of energy drinks in order to find the maximum absorption level. They can prepare a solution con-

taining the approximate caffeine concentration claimed by the manufacturer. Because of absorbance saturation, they will have to dilute the solution. They should decide to work at 271 nm, because there is an absorption peak at this wavelength.

- They can then produce a calibration curve with various aqueous solutions of caffeine and they can test it on a chosen energy drink diluted 20 times.
- Using this method they can deduce that the energy drink contains 17 % more caffeine (373 mg/L) than is claimed (320 mg/L) by the manufacturer. Of course the manufacturer did not cheat on the figures, because it has internal and external quality control procedures. However, the second compound that was found by the chromatography (vitamin B6 and/or B3), which also absorbs in the UV region, has an effect on their calibration curve.

FIG. 9 Caffeine absorption spectrum

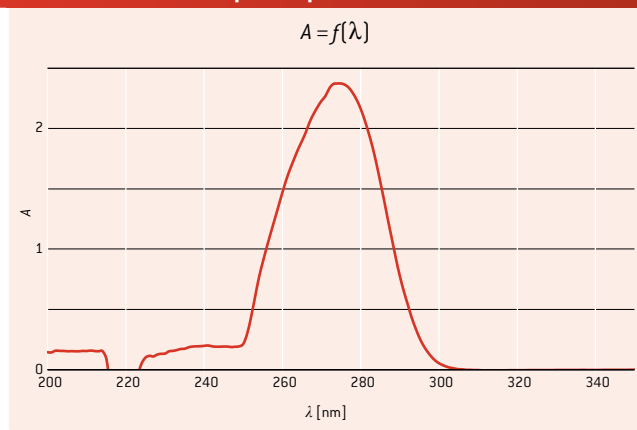
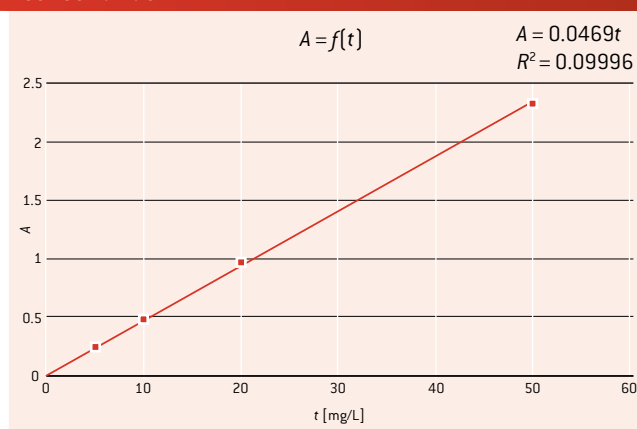


FIG. 10 Calibration curve of absorption linked to caffeine concentration



To get a better calibration curve:

- The students can produce the absorption spectrum of vitamin B6 and/or B3 to determine whether they absorb strongly at the wavelength that was previously chosen. According to the result, they can decide to choose another wavelength. Now that they have the spectra of B6 and B3, they can choose a wavelength at which the absorption is low (for example, between 240 and 250 nm).

FIG. 11 Absorption spectrum of vitamin B6

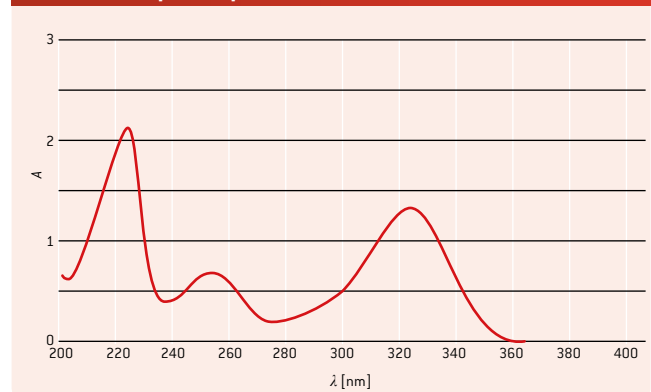
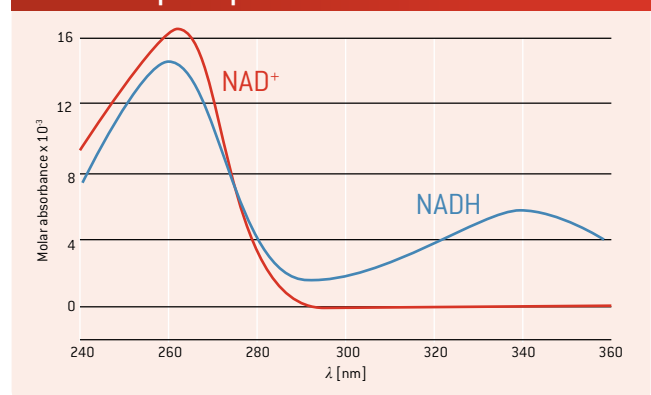


FIG. 12 Absorption spectrum of vitamin B3<sup>[1]</sup>



- It would also be very interesting to motivate the students to find another method of analysis such as HPLC in a laboratory; this would enable them to obtain a better result.

### 3 | 2 How to measure the effect of isotonic drinks and water on brain activity

Our bodies need water, sugar and minerals in order to work well. You can see a very impressive demonstration of that in a video of Gabriela Andersen-Schiess at the Olympic marathon in 1984, who failed to take a drink at the last beverage station. You can find several videos of this on the Internet.

We will develop methods, design a study and think about objectivity, validity and reliability as we measure the effect of isotonic drinks and water on brain effectiveness.

#### Biology:

Students of all ages should begin by pooling their knowledge. Students of age 13+ might go on to do research on various brain activities (sensors, actors, modal and intermodal activities, etc.) and the influence of water and isotonic drinks. Then they can present their results on posters before they start thinking about how to measure the effect mentioned above.

FIG. 13 Example of table for digit symbol substitution test

1	2	3	4	5	6	7	8	9
<	∩	Δ	X	+	⊥	∧	○	=
2	1	5	4	7	6	9	3	8
∩	<							
6	3	1	2	6	7	3	9	2

They may choose the following methods:

**[A] Number-symbol test (which is part of many IQ tests)—  
advised for students age 13+**

This test, which is also known as the digit symbol substitution test (DSST), helps to assess whether the subject has normally functioning intermodal activity.

On a piece of paper there is a list of numbers from, e.g., 1 to 9. Each number is associated with a symbol (e.g., - / & / 0). Under this list is a table with a list of numbers repeated in random order. The subject has to put the associated symbol beneath every number as fast as possible.

A student from the subject group can be given, e.g., 90 seconds to complete the sheet of paper. At halftime, e.g. 45 seconds, he or she makes a pause. You can check later whether the student is getting faster at associating numbers with symbols. This is the kind of brain activity that is called learning.

Five minutes later the student could be asked to write down the correct symbols associated with the numbers, in order to see how much he or she remembers. This is another kind of brain activity known as long-term memory.

**[B] Ruler test—recommended for all ages**

The test administrator lets a ruler fall down between the thumb and forefinger of the subject, and the subject tries to catch it as fast as possible. The students may discuss what the best starting position for the ruler would be. They can find out quite easily how far the ruler must fall before the subject can catch it.

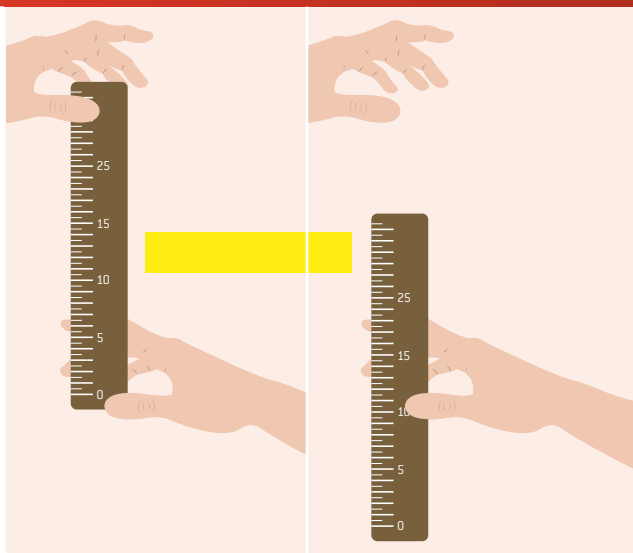
Moreover, they have to find out the best design for their study, including the time needed by a student who has not consumed any drink. This is of course an experimental control design, which means that you are simultaneously comparing two random groups (a control group and an experimental group). This setup makes it possible to compare the brain activity of two groups without any further influences or confounders apart from the factor of drinking. In further tests, the students can measure and compare the effects of different kinds of drinks.

**Maths:**

**[for test A]** The students (age 13+) will collect and analyse data and present what they have found out.

**[for test B]** The students will have to do some (mental) calculation in order to find out how many centimetres the ruler has fallen if they do not set the starting position of the subject's thumb at 0 cm. The youngest pupils might simply compare single results, whereas older ones might make calculations that take into account the uncertainties of measuring and then find the average of several measures.

FIG. 14 Ruler test



**Physics:**

**[for test B]** Students age 13+ might calculate the time during which the ruler was falling by using the height  $h$  they have measured.

$$E_{kin(1)} + E_{pot(1)} = E_{kin(2)} + E_{pot(2)}$$

$$E_{kin(1)} + 0 = 0 + E_{pot(2)}$$

$$\frac{1}{2} \cdot m \cdot v^2 = m \cdot g \cdot h \quad | : m$$

$$\frac{1}{2} \cdot v^2 = g \cdot h$$

with  $v = g \cdot t$  because  $v = a \cdot t$  and  $a = g$

$$\frac{1}{2} \cdot a^2 \cdot t^2 = g \cdot h \quad | \frac{2}{g^2}$$

$$t^2 = 2 \cdot \frac{h}{g} \quad | \sqrt{\quad}$$

$$t = \sqrt{2 \cdot \frac{h}{g}}$$

$a$ : acceleration [ $\frac{m}{s^2}$ ]

$h$ : height [m]

$g$ : gravitational acceleration,  $g = 9.81 \frac{m}{s^2}$

$t$ : time [s]

$v$ : velocity [ $\frac{m}{s}$ ]

Finally, you are welcome to share the results you have found in cooperation with other schools. You find further information on our website. <sup>[2]</sup>

**REFERENCES**

<sup>[1]</sup> Source: Cronholm144 [own work] [public domain], via Wikimedia Commons [https://en.wikipedia.org/wiki/Nicotinamide\\_adenine\\_dinucleotide#/media/File:NADNADH.svg](https://en.wikipedia.org/wiki/Nicotinamide_adenine_dinucleotide#/media/File:NADNADH.svg) (08/03/2016)

<sup>[2]</sup> [www.science-on-stage.de/iStage3\\_materials](http://www.science-on-stage.de/iStage3_materials)

**4 | CONCLUSION**

This project is adaptable and can be used to teach students ranging from ages 8 to 18 how to measure brain activity and how to optimise a method in order to minimise the need for evaluation by calculation, counting etc. Students will get to know the experimental control design and are welcome to bring in STEM aspects they have learned in biology, maths or physics.

**5 | COOPERATION OPTIONS**

We advise you to think about this project as an interscholastic and international project. If you do not have the required technical equipment for the chemistry part at your school, you could contact other schools nearby so that you can cooperatively conduct experiments with them. Your pupils will have to communicate their investigations and protocols to the other pupils; this makes much more sense for them than just writing down their results in their exercise books. This kind of cooperation and sharing generates further motivation and input and adds a bilingual option to teaching/learning STEM subjects.

You can compare drinks that are available in different countries and the attitudes toward consuming them. You can also discuss the design of the studies, gather more ideas and do the exercises at two or more cooperating schools in order to get more data for your analysis of the effects.



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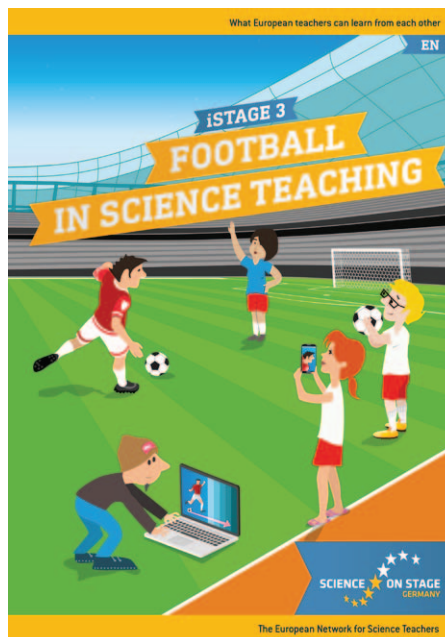
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