A negative test is not no sugar

Sucrose is a non-reducing sugar: it does not give a positive Benedict's test directly. Hydrolysis breaks it into reducing sugars that can give a positive result.

SUCROSE HYDROLYSIS: A WORD-EQUATION MODEL

sucrose + water

↓ dilute acid + heat

glucose + fructose

One sucrose molecule uses one water molecule to form one glucose and one fructose molecule. Both products are reducing sugars.

1. WHAT NON-REDUCING MEANS

In the standard test, sucrose cannot reduce the copper(II) ions in Benedict's reagent. Its glycosidic bond involves both anomeric carbons, so there is no free reducing end. This does not mean sucrose cannot be digested or used as an energy source.

2. WHAT HYDROLYSIS CHANGES

Hydrolysis breaks the glycosidic bond using water; dilute acid catalyses the reaction and heating speeds it up. The products can react in Benedict's test. The acid does not itself create the coloured Benedict's precipitate.

3. THE BEFORE-AND-AFTER LOGIC

A negative direct test followed by a positive test after hydrolysis supports the presence of a non-reducing sugar in a simple sugar sample. It is the change in test response that matters. The final test detects reducing sugars produced by hydrolysis.

Scope: Cambridge Biology 9700 syllabus, section 2.1, outcome 3, p. 17.

Test, hydrolyse, then retest

Use separate portions of the original sample. Do not add acid to the tube already used for the first Benedict's test: it contains reagent and reaction products.

1. DIRECT BENEDICT'S TEST

Add Benedict's solution to one portion and heat in a hot water bath. Record whether blue remains or a green, yellow, orange or brick-red positive result develops.

2. HYDROLYSE A FRESH PORTION

To an untreated portion of the same sample, add dilute hydrochloric acid and heat using the approved water-bath procedure. This can split sucrose into glucose and fructose.

3. COOL AND NEUTRALISE

Allow the mixture to cool. Carefully add the prescribed alkali, such as sodium hydrogencarbonate, to neutralise the acid; check with pH paper as instructed. Add gradually because fizzing can occur.

4. ADD FRESH BENEDICT'S REAGENT

Once the acid has been neutralised, add Benedict's solution. Its alkaline conditions are needed for the reducing-sugar reaction; residual acid can interfere and cause a misleading result.

5. HEAT AGAIN AND RECORD

Heat in the hot water bath and observe. A positive result after an initially negative direct test is evidence that hydrolysis produced reducing sugars. Compare the two recorded observations.

Two heating stages in the treated portion: first with acid for hydrolysis, then with Benedict's reagent for detection. Neutralisation comes between them.

Safety: wear eye protection, handle hot tubes with holders and avoid acid or alkali splashes. Do not seal a fizzing tube. Follow the teacher's quantities, timings and disposal rules.

Method order corroboration: Cambridge 9700/13, June 2017, Q5 (mirror).

Interpret both observations

The direct test checks for reducing sugar already present. The treated test checks whether reducing sugar is detectable after hydrolysis and neutralisation.

DIRECT TEST	AFTER TREATMENT	WHAT THE PAIR SUPPORTS
Negative	Positive	In a simple sugar sample, non-reducing sugar is indicated: reducing sugars became detectable after hydrolysis.
Positive	Positive	Reducing sugar was already present. These two positive results alone do not establish whether non-reducing sugar is also present.
Negative	Negative	Neither test detected reducing sugar under the conditions used. This does not prove that all carbohydrates are absent.
Positive	Negative	An unexpected outcome. Check the method, dilution, neutralisation, heating and reagents before drawing a conclusion.

1. USE CONTROLS TO CHECK THE PROCEDURE

Check Benedict's reagent and heating with glucose. A known sucrose control should be negative directly and positive after treatment. Carry a water blank through the full procedure to detect contamination.

2. LIMIT THE CLAIM IN COMPLEX SAMPLES

Interpretation caution: starch and other hydrolysable carbohydrates can also yield reducing sugars. The test is not uniquely specific to sucrose; consider sample composition and other tests before naming a molecule.

Scope: Cambridge Biology 9700 syllabus, section 2.1, outcome 3, p. 17.