Enzymatic kit for determination of Sucrose + D-Glucose + D-Fructose

OENOLAB

Principle:

Sucrose + H_2O $\xrightarrow{INV}$ D-glucose + D-fructose

D-Fructose + ATP $\xrightarrow{HK}$ Fructose-6-Phosphate + ADP

Fructose-6-Phosphate $\xrightarrow{PGI}$ Glucose-6-Phosphate

D-Glucose + ATP $\xrightarrow{HK}$ Glucose-6-Phosphate + ADP

Glucose-6-Phosphate + $NADP^+$ $\xrightarrow{G6PDH}$ D-Gluconate-6-Phosphate + NADPH + H^+

Kit for 45 analyses includes:

Bottle	Composition	Quantity
A.	Buffer	100 ml
B.	Enzymes	13.6 ml
C.	Invertase	20 ml
STD.	Standard 10 g/l	2 ml

Reagent preparation:

R1: is **Bottle A** and it's ready to use.

R2: is **Bottle B** and it's ready to use.

Invertase solution: Mix 1 volume of **Bottle C** with 1,5 volume of distilled water. Stability of invertase solution: 1 month at 2 to 8°C.

Sample preparation:

If necessary, filtrate or centrifuge the sample in order to eliminate the particles in suspension.

In a tube, mix together 1 ml of sample and 1 ml of invertase solution. Leave the tube at room temperature for 1 hour.

Performance:

This kit has been developed to determine the total concentrations of Sucrose, D-Glucose and D-Fructose in a measuring range from 0 to 10 g/l. If the expected values are higher than 10 g/l, samples should be diluted with distilled water and the result must be multiplied by the dilution factor.

Storage instructions and reagent stability:

The reagents are stable up to the expiry date, if stored at 2 to 8°C. Contamination should be avoided. Do not freeze the reagents!

Warnings and precautions:

Do not swallow the reagents. Avoid contact with the skin and mucous membranes.

Take the necessary precautions for the use of laboratory reagents.

Sample analysis:

	Blank	Standard	Sample
R1	2200 µl	2200 µl	2200 µl
Water	30 µl		
Standard		30 µl	
Sample			30 µl
Mix and read	**DO1 blank**	**DO1 standard**	**DO1 sample**
R2	300 µl	300 µl	300 µl
Mix wait 15 min and read	**DO2 blank**	**DO2 standard**	**DO2 sample**

Wavelength: 340 nm.

Cuvette: 1 cm light path (plastic or glass)

Temperature: 20 – 37°C

Zero against water or air

Calculation:

ΔDO sample = (DO2-DO1) sample - (DO2-DO1) blank

ΔDO standard = (DO2-DO1) standard - (DO2-DO1) blank

$$C \text{ sample (g/l)} = C \text{ standard} \times \frac{\Delta DO \text{ sample}}{\Delta DO \text{ standard}}$$

Dilution factor of the sample has to be considered in the calculation