iCheck **Carotene:**
Measurement Step-by-step

BioAnalyt GmbH

Outline

1. Introduction to iCheck Carotene
2. Using iCheck Carotene Step by Step
3. Handling and Disposal

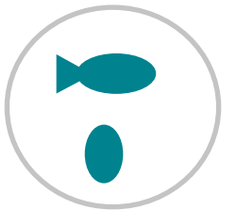
iCheck Carotene is a Portable All-Inclusive Test Kit

- iCheck Carotene measures carotenoids in vitamin premix, food and biological fluids
- It is a portable single-wavelength photometer, precalibrated for quantitative measurement of carotenoids.
- Carotene reagent vial has a precise volume and ratio of organic solvents for optimal extraction of carotenoids from the sample.

iCheck Carotene Measures Carotenoids in Food and Biological Fluids

- iCheck Carotene validated to measure carotenoids in following samples:

Beverages



Salmon, eggs

Vitamin Premix

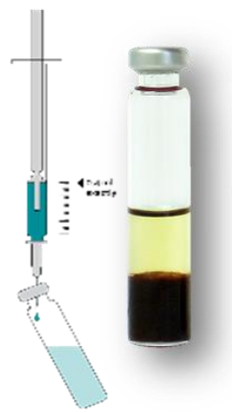
Cattle Blood

Roots (i.e. cassava)

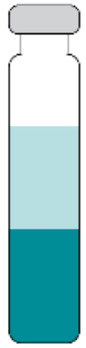
Breast Milk

Science Behind iCheck Carotene

Natural or added carotenoids



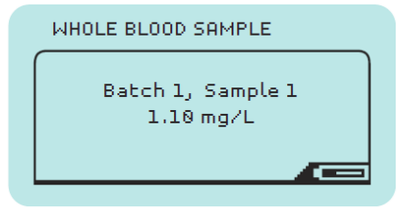
Extraction of carotenoids into the upper organic phase



Separation of phases

450 nm

iCheck Carotene measures absorption of 450 nm by carotenoids in a sample.



Software converts the intensity of light emission into mg/L.

iCheck Carotene is Precalibrated and Comes with a 2-year Warranty

- iCheck Carotene is pre-calibrated during production. Therefore, no user calibration is necessary.
- iCheck Carotene comes with a 2-year warranty.
- The calibration of the device is controlled on 2 levels:
  - Automatic Self-Test
  - Device Control with Carotene Standard
- The shelf-life of the reagent vials is 1 year at 20-30 °C.

Provitamin A carotenoids and conversion to vitamin A

USA and Canada

1 mg Retinol Activity Equivalent (RAE)

- = 1 mg retinol
- = 2 mg supplemental beta-carotene
- = 12 mg dietary beta-carotene
- = 24 mg other dietary provitamin A carotenoids

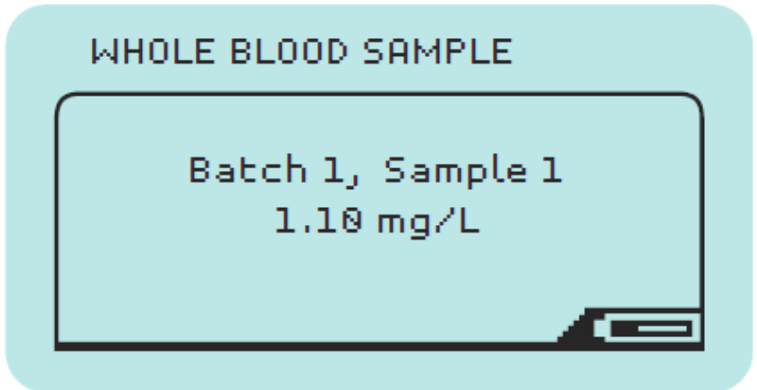
Europe

1 mg Retinol Equivalent (RE)

- **= 1 mg retinol**
- **= 6 mg beta-carotene**
- **= 12 mg other provitamin A carotenoids**

If you need support with conversion, you can use Units Converter available on iCheckAcademy.org

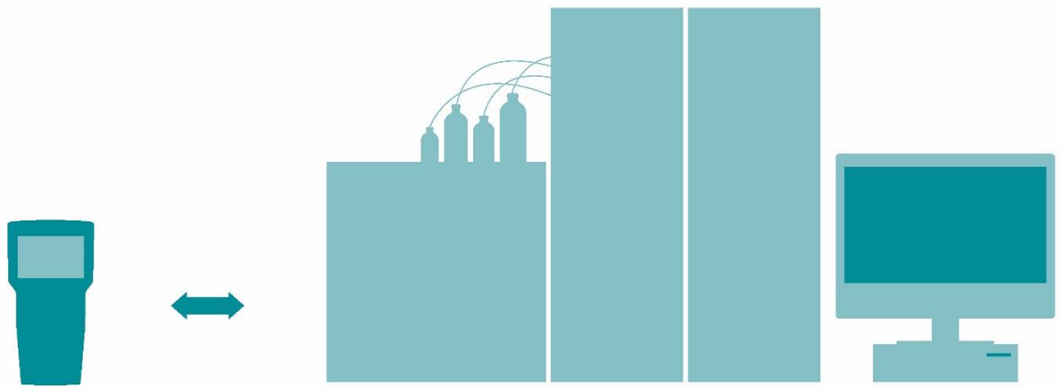
iCheck Carotene Measurement Range is 0.15 – 15.0 mg/L

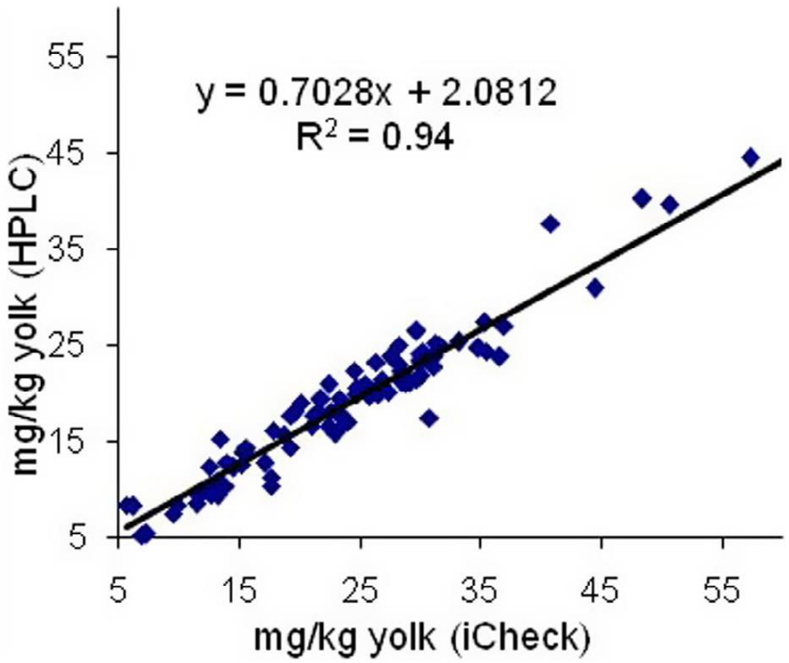


- iCheck Carotene results are displayed in mg per litre and indicates the total carotenoids concentration in the sample
- iCheck Carotene measurement range is:
  - 0.15 – 15.0 mg/L

Measurement Limit	Device Display	Observed in the vial
< 0.15 mg/L	Measured Value	No yellow color
> 15 mg/L	Value above 15 mg/L	Intense yellow color

iCheck Carotene is validated and the results published in peer reviewed journals

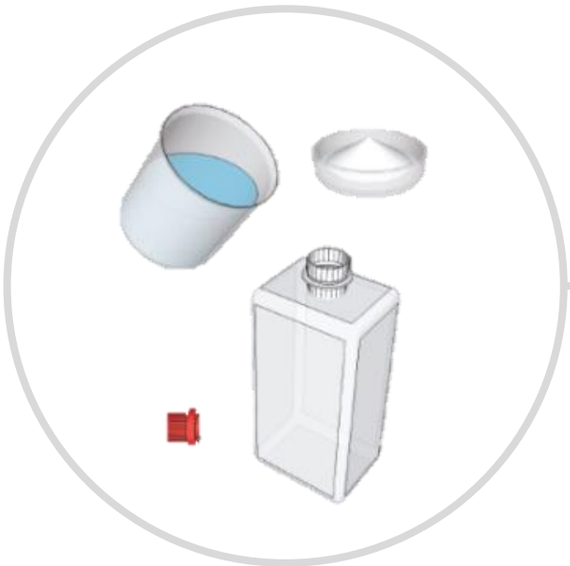




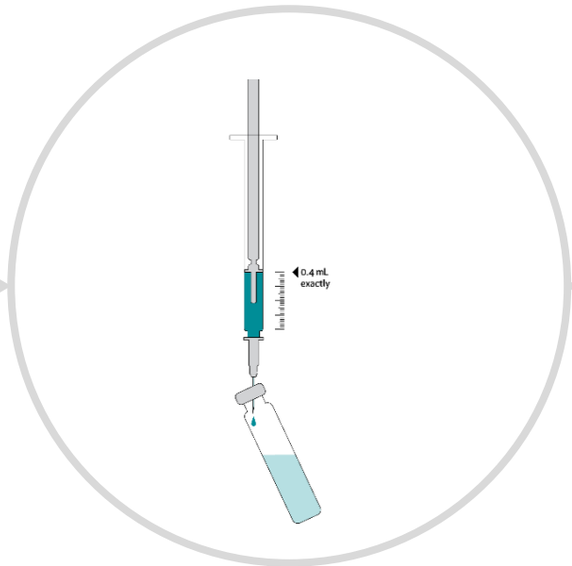
- Comparison to the reference method of HPLC yields correlation coefficient of **0.94 to 0.99**.
- Coefficient of variation in cattle blood is **3.5%**.

- *References:*
  - *Schweigert FJS. Determination of ß-carotene in whole blood of cattle Comparison of a new cow-side assay with HPLC. Vet. Clin. Path., 2011.*
  - *Islam KMS, Schweigert FJS. Comparison of three spectrophotometric methods for analysis of egg yolk carotenoids. Food Chemistry 172, 233–237, 2015.*

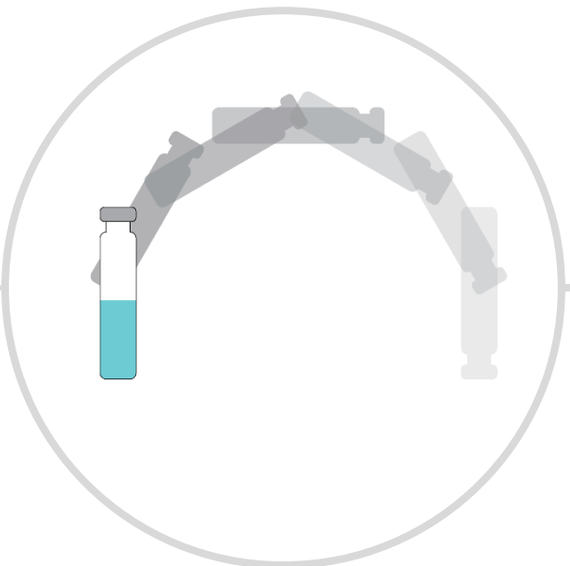
iCheck Carotene Measures in a 4-step Procedure



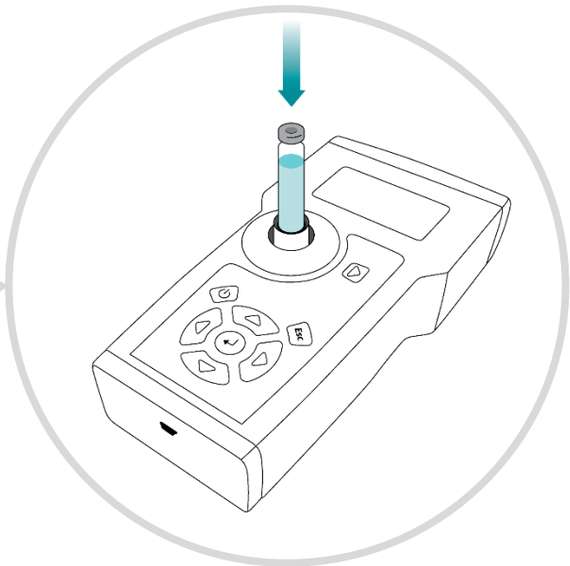
1. Sample Preparation



2. Injection

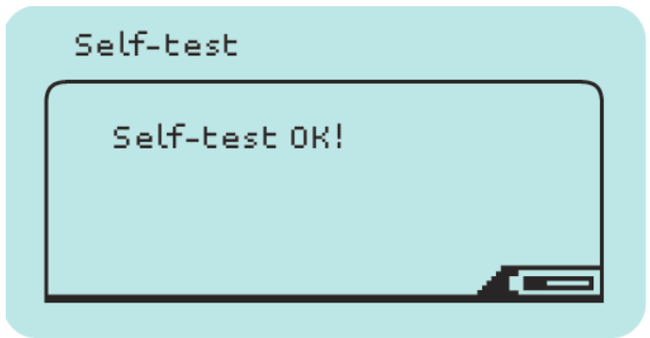


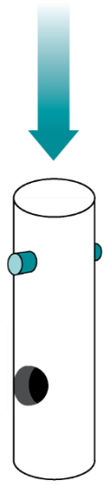
3. Reaction



4. Measurement

Step 1: Device Control





Attention

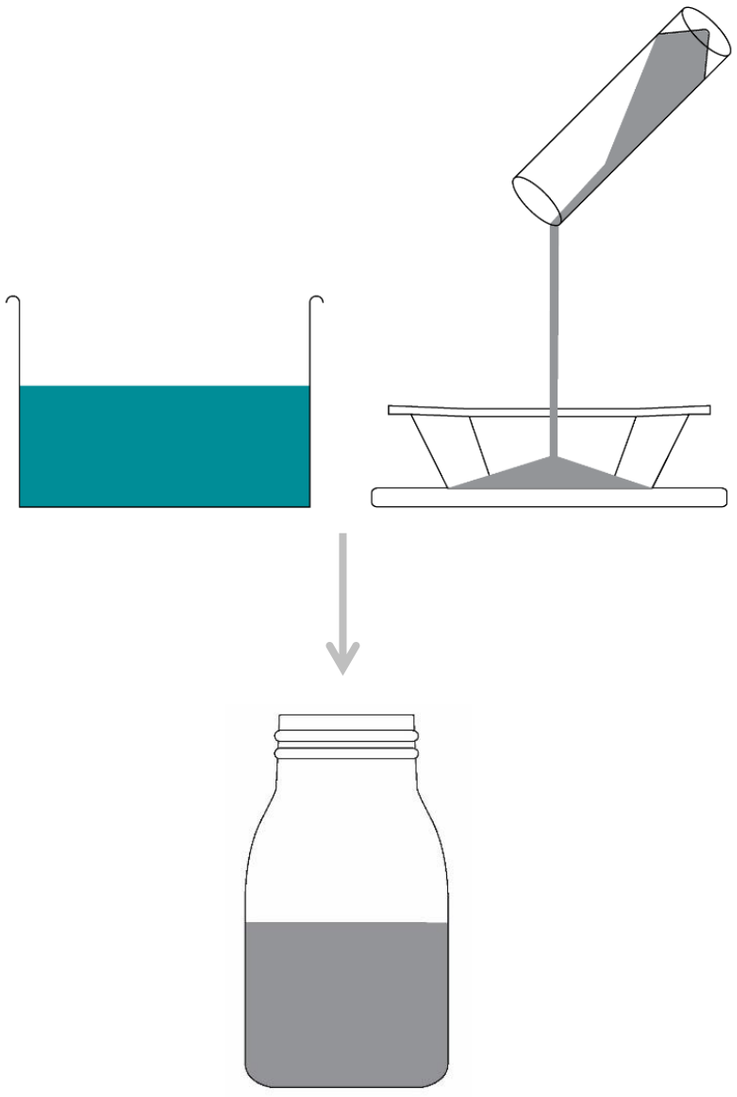
- Do not measure outside the temperature range of 20 – 30 °C
- Do not allow any dirt or dust to enter the measurement chamber

Control your iCheck Carotene:

- Measurement must be done on a flat surface. Keep the metal cap on the measurement chamber.
- Turn on your iCheck™ Carotene. It will run automatic **self-test**.
- Insert Carotene Standard into the measurement chamber.
- Go to **Device Control** measurement mode and press measurement key.
- Control the value displayed against the range indicated on the standard case.
- *Control your device before each set of measurements*

Step 1b: Sample Preparation

For more detailed instructions on specific sample preparation protocols refer to the "iCheck Carotene_Sample Preparation" file.



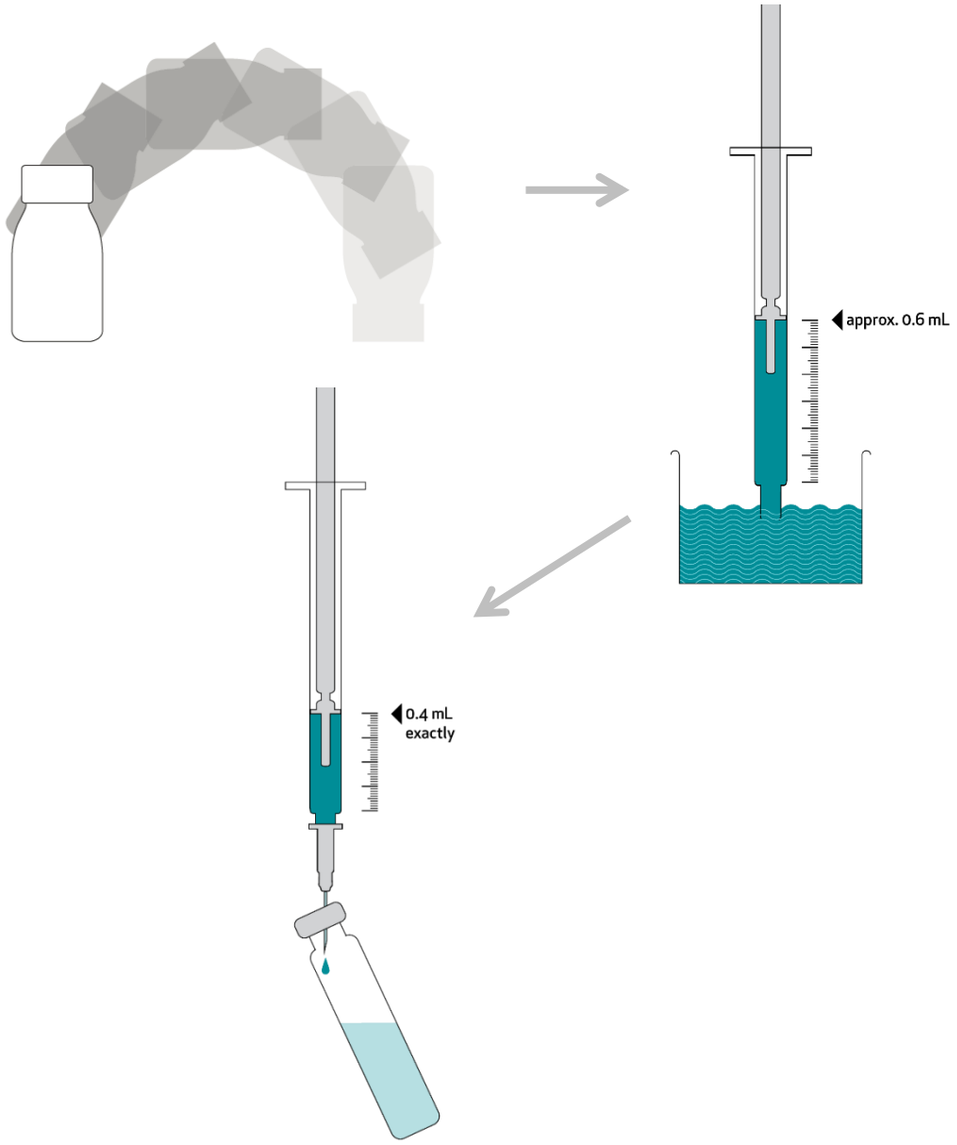
- Prepare the sample by diluting it with water.
- Sample preparation should be done if

1. expected concentration of your sample is above iCheck Carotene measurement range (> 15.0 mg/L)
2. your sample is a solid sample.

Important!

- Total Carotenoids concentration of the sample solution must be in the measurement range of iCheck Carotene which is 0.15 – 15.0 mg/L.

Step 2: Injection



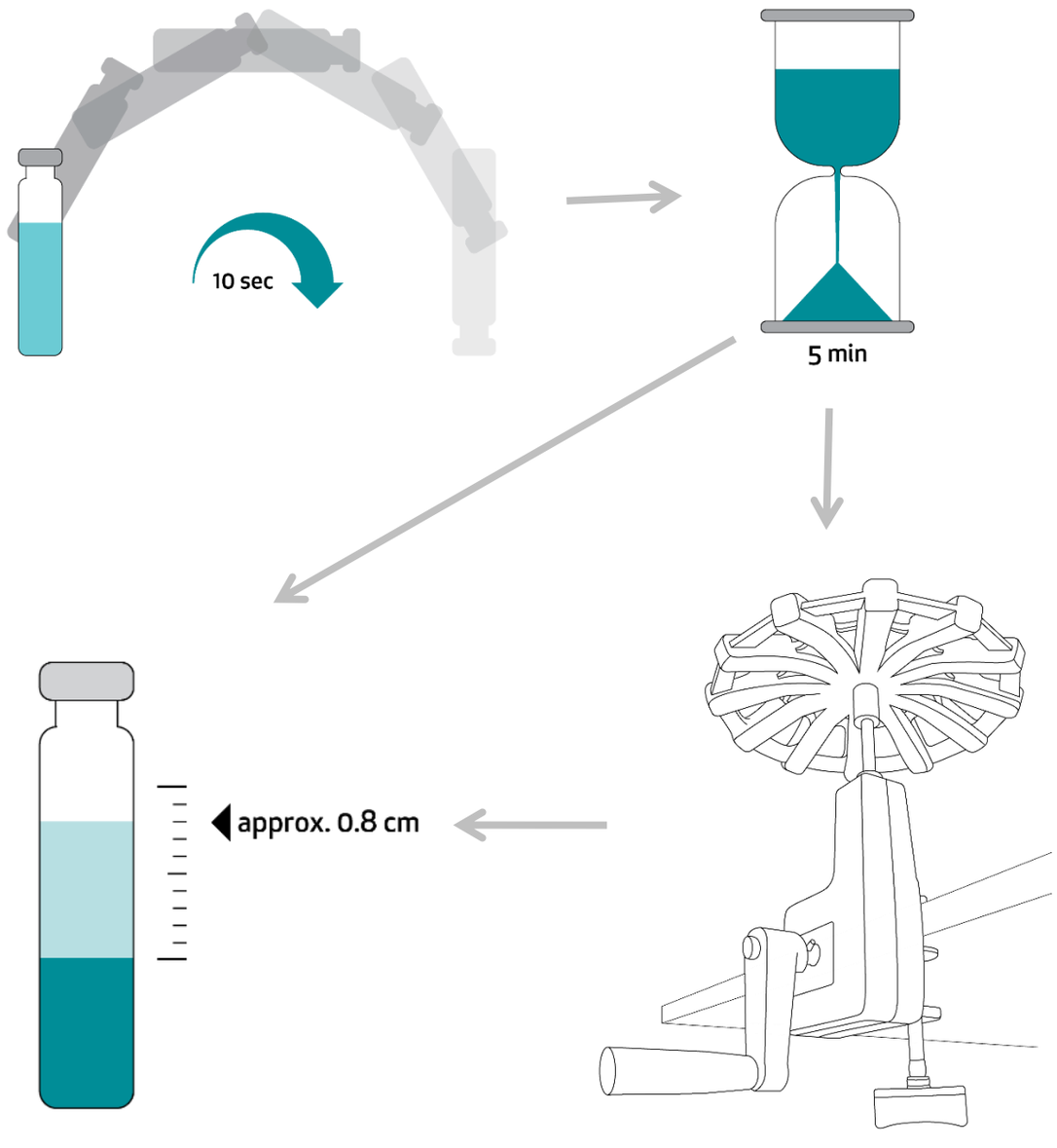
Inject the diluted sample

- Vigorously shake the sample solution to homogenize.
- Quickly take up approx. 0.6 mL of the sample solution with the syringe.
- Put on the needle and make sure no air bubbles are in the syringe.
- Adjust the volume to exactly 0.4 mL into paper tissue.
- Inject 0.4 mL sample into the reagent vial.

Attention

- The sample must not settle down in the solution
- There should be no air bubbles in the syringe

Step 3: Extraction



Extraction of total carotenoids:

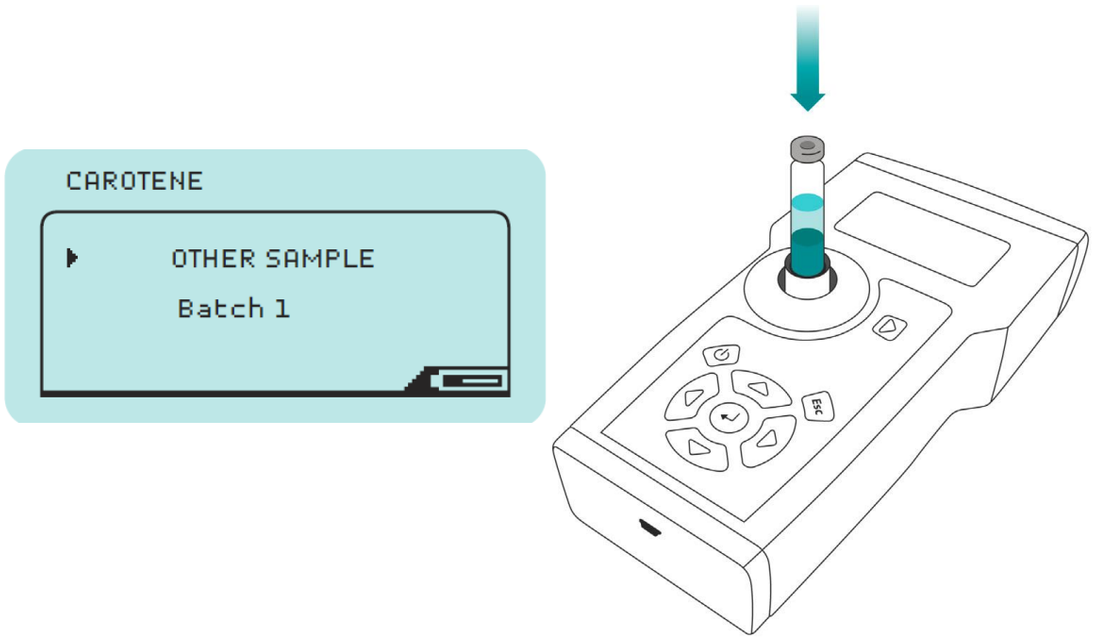
- Vigorously shake the vial for 10 seconds. The content of the vial should appear as one uniform solution.
- Let the vial stand still for 5 minutes until the solution in the vial appear as two distinct phases.
- Make sure nothing is stuck on the inside of the glass in the vial on the level of the upper clear phase. If a piece of sample is stuck on the inside tap gently the vial on the solid surface to shake off the piece and wait again for phase separation.

Note: some samples do not separate into 2 phases. To facilitate this use BioAnalyt centrifuge.

Attention

- Do not measure a reagent vial with NO clear supernatant of at least 0.8 cm (see picture)
- Do not store vial in the fridge

Step 4: Measurement



Attention

- Do not measure in the direct sunlight
- Always measure in the temperature range of 20 – 30°C

Measure the reagent vial with the sample:

- Make sure the vial is clean. Wipe the glass surface with paper tissue
- Set the iCheck Carotene to <u>OTHER SAMPLE</u> mode.
- Press measurement key and follow the instructions on the display.
- iCheck™ Carotene will display the result in mg/L. This value indicates the concentration of total carotenoids (TC) in the injected sample solution.
- The vial with the sample must be measured with iCheck earliest 5 minutes and latest 1 hour after sample injection.

Calculations of Dilution Factor and Measured Concentration

1

Dilution Factor (DF) =
Total Diluted Sample Volume [mL] / Sample [g]

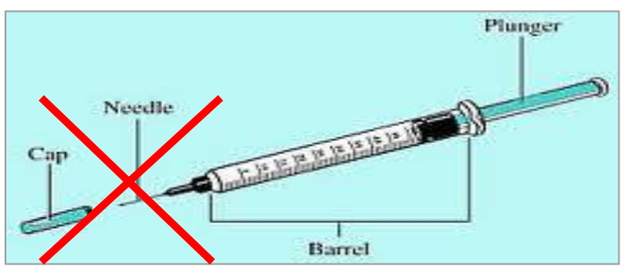
2

Measured Total Carotenoids [mg/kg] =
iCheck Carotene result [mg/L] x DF

Disposal Instructions for Vials and Syringes

REAGENT VIALS

Dispose of vials as hazardous material according to local regulations and upon consultation with local waste disposal services. Handle any broken glass using protective gloves.



SYRINGES

Take extra care when handling syringes with needles. DO NOT RECAP the needle!

Discard the needles into specific containers.

For Technical Support email:

support@bioanalyt.com

or WhatsApp:

+49 332 835 150 034

Contact for orders:

E-mail: contact@bioanalyt.com

www.bioanalyt.com

Follow us on:

BioAnalyt

measure for life