# iCheck Carotene Measuring Carotenoids in Cassava Roots

## **1. Method Principle and Application**

iCheck Carotene is a portable, allinclusive test kit designed for the quantitative determination of total carotenoid content food and in biological samples. The method is based on the light absorption of carotenoids at 450 and 525 nm. Using an integrated algorithm, the device calculates the carotenoid concentration in mg/L. The measurement range of iCheck Carotene is 0.15 to 15 mg/L.



## 2. Carotenoids in Cassava and Biofortified Cassava

Vitamin A deficiency (VAD) is a major public health issue in many low- and middle-income countries (LMICs), particularly affecting preschool-age children and pregnant women. Globally, an estimated 190 million preschool-age children (approximately 30%) and over 19 million pregnant women suffer from VAD, leading to increased risk of morbidity, impaired immune function, and blindness. In 2013 alone, an estimated 105,700 deaths among children under five in LMICs were attributed to VAD (Black et al., 2013).

Cassava (*Manihot esculenta*) is a key staple crop in sub-Saharan Africa, Latin America, and parts of Asia, providing daily calories for over 500 million people. In Nigeria alone, more than 100 million people consume cassava daily, often as a primary energy source. Global cassava production reached 278.7 million tons in 2015 and rose to approximately 288.4 million tons in 2016 (FAO, 2017).

To address VAD through cassava, biofortification efforts have aimed to increase provitamin A carotenoid (pVAC) concentrations in cassava roots. Non-biofortified cassava varieties typically contain negligible amounts of carotenoids (~0 ppm), whereas biofortified varieties target levels of up to 15 ppm fresh weight (30 - 35 ppm dry weight) total carotenoids, which can contribute significantly to daily vitamin A intake, especially considering the 20% estimated bioavailability of carotenoids from cassava.

The development of vitamin A-biofortified cassava involves conventional cross-breeding strategies combined with phenotyping and, more recently, molecular marker-assisted selection and high-throughput genotyping. Selection for high-carotenoid lines starts with screening breeding populations using both visual and analytical methods. While root color (white to yellow) serves as a basic qualitative indicator of carotenoid presence, it is insufficient for accurately distinguishing between yellow-root varieties with different carotenoid concentrations.

Therefore, quantitative analytical methods, such as portable spectrophotometry (e.g., iCheck Carotene), are employed to measure total carotenoid content more precisely. This tool enable breeders to reliably identify and select genotypes that meet or exceed target carotenoid levels, thereby accelerating the development of nutritionally improved cassava varieties.

#### References

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- Saltzman, A. et al. (2013). Biofortification: Progress toward a more nourishing future. Global Food Security, 2(1), 9–17.
- HarvestPlus (n.d.). Vitamin A Cassava. HarvestPlus. Retrieved June 20, 2025, from <u>Vitamin A Cassava – HarvestPlus</u>.
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## 3. Analyzing Carotenoids in cassava roots with iCheck Carotene

- Wash and peel the root. Cut ~0.5 cm pieces from different parts, then mix well (image 1).
- Weigh 5 g and grind in a mortar to a smooth paste (images 2 & 3).
- Add 5 mL distilled or bottled water, mix thoroughly (image 4).
- Transfer the paste to a 50 mL Falcon tube; rinse the mortar with 15 mL water and add the rinse to the same tube (image 5).
- Immediately draw the well-mixed slurry into a syringe and inject it into the iCheck Carotene vial (image 6), following the instructions in the <u>iCheck Carotene User Manual</u>.



## 4. Dilution Factor (DF) Calculation

To obtain the carotene concentration of the original wet cassava root sample, you must first calculate the dilution factor according to the following formula:

$$DF = \frac{Total \ sample \ solution \ volume \ (mL)}{Sample \ weight \ (g)}$$

Once you have calculated the dilution factor, multiply the iCheck carotene result by the dilution factor.

Carotene in cassava 
$$\left(\frac{mg}{kg}\right) = iCheck Carotene result \left(\frac{mg}{L}\right) x DF$$

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