

iCheck Iron

Measuring Iron in Fortified Rice Kernels

1. Method Principle and Application

The [iCheck Iron device](#) is a single-wavelength portable photometer that converts absorption units into iron concentration in mg per liter (mg/L). The reagent vials supplied in the iCheck Iron Test Kit are developed to extract iron from water soluble samples and detect iron using colorimetric reaction.



Acidic environment reduces ferric iron (Fe^{3+}) to ferrous iron (Fe^{2+}) which reacts with bathophenanthroline. The resulting red complex of iron and bathophenanthroline correlates with iron concentration and is measured at 525 nm with the iCheck device. The measurement range of iCheck Iron is 1.5 – 12.0 mg/L.

The method is suitable for iron quantification in vitamin premixes, fortified rice kernels (FRK), flour, corn soya blend (CSB), lipid-based nutritional supplements (LNS), soy and fish sauce and beverages. Handling requires only one day training.

2. Working with Highly Concentrated Samples

If the expected iron concentration is outside of the iCheck device measurement range, the samples must be diluted, ideally to fit the middle of the measurement range. Highly concentrated samples should be handled with extra care due to a higher chance of error when working with low weights and small volumes.

Small errors lead to large variation!

Tips for accurate results

- Ensure your balance is well-calibrated.
- Record the exact amounts of samples weight and diluted volume for dilution factor calculation.
- Ensure your diluted sample is homogenous.

3. Iron in FRK

Typically, iron is added to fortified rice kernels (FRK) as ferric pyrophosphate (FPP) or sodium iron ethylenediaminetetraacetate trihydrate (NaFeEDTA). The protocol detailed in this document can be used for either type of iron, as it is suitable for iron compounds that are insoluble and soluble in water.

4. iCheck Iron Performance with FRK

iCheck Iron performance is assessed following a standardized process that combines assessment of precision, trueness and a comparison to a reference method (i.e. atomic absorption spectroscopy). The detailed description of this process is provided in the iCheck Iron Performance Guide. Performance of iCheck Iron with FRK has been assessed in internal validations. Below is a table detailing the observed precision and recovery.

Table 1. iCheck Iron Performance with FRK

Sample Type	Recovery	Coefficient of Variation
FRK	97 – 102%	Max. ±5%

5. Analyzing Iron Content in FRK with iCheck Iron

A. Prepare NaOH and HCl Solutions

0.6% NaOH:

- Dissolve 6 g NaOH in 1000 mL distilled water
- NaOH must be added to water, not the other way around.
- The solution is corrosive.
- The solution is enough for approximately 40 sample preparations and is stable for 6 months at room temperature.

0.2 M HCl:

- Dilute 7 mL of concentrated HCl (~36%) with 400 mL of distilled water.
- HCl must be added to water, not the other way around.
- The solution is corrosive.
- The solution is enough for approximately 40 sample preparations and is stable for 6 months at room temperature.



B. Prepare FRK for Analysis

Based on iron content in the FRK, sample weight and volumes should be adapted (see Table 2).

Table 2. Dilution of FRK for Iron Measurement with iCheck Iron

	Expected Iron Conc. in FRK [mg/kg]	Sample Weight (finely ground FRK) [g]	Volume of FRK Slurry	Final Dilution	Expected iron conc. in diluted sample, [mg/L]
B	1 000 - 5 500	1.0	0.5 mL	1: 525	1.9 – 10.5
C	5 500 - 10 000	0.5	0.5 mL	1: 1050	5.2 – 9.5
D	1 300 - 5 500	0.5	1.0 mL	1: 550	2.4 – 10.0

Only use dilution procedure D if the FRK slurry turns too viscous during the incubation in NaOH using dilution procedure B.

Step 1: Prepare FRK Slurry

- Grind approx. 20 g of fortified rice kernels (FRK) into fine flour-like powder using a coffee grinder. Make sure no large particles are left.
- Transfer 25 mL of 0.6% NaOH solution to a 50 mL Falcon tube.
- Weigh in 1.0 g or 0.5 g of FRK powder, based your expected iron concentration (Table 2) and add it to 25 mL of NaOH solution. Shake immediately for 30 seconds to create FRK slurry.
- Incubate for 30 minutes at room temperature. Shake for 30 seconds every 10 minutes.

Step 2: Make Final Dilution in HCl

- Transfer 10 mL of 0.2 M HCl solution to a 15 mL Falcon tube.
- Shake the FRK slurry and quickly take up 0.5mL or 1mL with the syringe or a pipette.
- Add the 0.5mL or 1mL of FRK slurry to 10 mL of 0.2 M HCl solution.
- Shake the mixture for 10 seconds and let it stand for 5 minutes.

C. Measure Iron Content in FRK

- Proceed with the measurement following the instructions in the [iCheck Iron User Manual](#).



6. Dilution Factor (DF) Calculation

The value displayed on the iCheck Iron after measurement will reflect the concentration of iron in the diluted sample.

To obtain the original FRK iron concentration, you must first calculate the dilution factor according to the following formula:

$$\text{Step 1} = \frac{\text{FRK sample weight (g)}}{\text{NaOH volume (mL)}}$$

$$\text{Step 2} = \frac{\text{FRK slurry volume (mL)}}{\text{HCl volume (mL)}}$$

$$\text{DF} = \text{Step 1} * \text{Step 2}$$

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

$$\text{Iron in FRK} = \text{iCheck Iron result} \left(\frac{\text{mg}}{\text{L}} \right) \times \text{DF}$$

For technical support email us: support@bioanalyt.com

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