Potentially eligible participants  
\( n = 7526 \)

Excluded  
\( n = 5074 \)  
- Systematic sampling of samples with normal red cell indices  
- No index test  
\( n = 261 \)  
- Age < 12yrs or > 19yrs  
- Missing demographic data

Index test  
\( n = 2273 \)  
(low MCV/MCH: \( n = 1686 \))  
(normal red cell indices: \( n = 587 \))

Hepcidin >3.2 ng/mL  
\( n = 1374 \)

No reference\(^1\) test  
\( n = 20 \)  
- Missing ferritin data

Iron deficient\(^2\)  
\( n = 1251 \)

Final diagnosis  
True negative  
\( (n = 1251/1354, 92.4\%) \)  
False negative  
\( (n = 123/1354, 9.1\%) \)

Hepcidin <3.2 ng/mL  
\( n = 899 \)

No reference\(^1\) test  
\( n = 50 \)  
- Missing ferritin data  
- Missing sTfR data

Iron replete\(^3\)  
\( n = 319 \)

Final diagnosis  
True positive  
\( (n = 319/849, 37.6\%) \)  
False positive  
\( (n = 580/849, 68.3\%) \)

\(^1\) MCV<80fL and/or MCH<27pg  
\(^2\) Subsequent ROC and Youden Index analyses identified a hepcidin value of 3.2 ng/mL as the cut off that optimally identifies iron deficiency in this population, predicting the highest sensitivity and specificity.  
\(^3\) Iron deficient defined by: Ferritin <15 ng/mL, or ferritin<30 ng/mL if CRP>5 mg/L, & sTfR-F>2