Objectives: The QuantiFERON-TB Gold in Tube (QFN) assay is now widely used for screening individuals at increased risk of developing tuberculosis (TB) such as close contacts to patients with active TB as well as planned immunosuppressive therapy. It has been assumed that the interpretation of QFN is more standardized and objective than the assessment of the tissue skin test (TST) induration. However, there is an increasing awareness that QFN results around the cut off (0.35 IU/ml) may cause difficulties in the interpretation such as within-subject variability. Our objective was to investigate the prevalence and impact of borderline reactive QFN results under routine clinical conditions. Methods: We included consecutive QFN samples (n=3785) from two laboratories in Sweden. For borderline reactive QFN results (defined as 0.2-0.99 IU/ml), a questionnaire was sent out together with an offer to submit another sample. Information about clinical symptoms, previous TST-testing, close contact to active TB or previous TB treatment as well as conditions while taking the blood sample was requested. Results: During the study period, 7.7 % (292/3785) of the samples were borderline reactive in the QFN test whereas 69.7 % (n=2637) were negative (<0.2 IU/ml), 20.0 % (n=756) positive (=>1.0 IU/ml) and 2.6 % (n=100) indeterminate. Among the 135 borderline reactive samples where a follow up test was submitted, 44.4 % (n=60) showed a persistent borderline reactivity, 46.7 % became negative (<0.2 IU/ml) and 8.9 % turned positive (=>1.0 IU/ml). Conclusion: This study suggests that for patients showing a borderline reactive QFN result (0.2-0.99 IU/ml) a new sample should be requested as there is a significant variability around the cut off which influences the clinical interpretation of the result.