Quantitative test procedure for small-scale mill fortification

All samples will be sent to a GAIN certified laboratory to perform a quantitative analysis assessing the uniformity/homogeneity of the fortification.

Test procedure is as follows:

a. **10 batches** (or "hoppers") of 1 bucket (~18kg) of grain will be milled/fortified separately.

b. From each batch, **6 individual samples** of fortified flour will be collected post mill (*sample collection procedure is described later*)

c. So, there will be a total of **60 samples** (300g each) of fortified flour collected.

In addition to the above, we will need to collect the following samples BEFORE the fortification:

a. Six samples of whole grain (non-milled Maize) will be collected to determine the native nutrient content (i.e. our control #1).

b. Additionally, before the “contamination” of the mill by the fortificant, 1 bucket (~18kg) of grain will need to be milled without fortification. 6 samples will be collected (sample collection procedure is described below). This will provide us with information about the nutrient content in the unfortified milled flour (i.e. our control #2).

The reason for collecting these unfortified milled flour samples is to determine if the native nutrient content differs from that of the non-milled whole grain samples. This will establish if some iron, for example, has been added into the flour during the milled process, potentially due to the hammer mill's metal components.
Labeling of samples:

All samples (i.e. 6 whole grain, 6 non-fortified flour and 60 fortified flour, each weighing approximately 300g) will need to be packed in zip-locked plastic bags and clearly labeled.

Labels will have 4 components: MILL - GRAIN - BATCH - SAMPLE

1. **MILL**: If we are undertaking the trials in more than one mill, then Mill IDs are labeled as A, B, C, etc.

2. **GRAIN**: The second component will represent Grain type, labeled as either C (corn/maize), M (millet), or W (wheat). For Tanzania we will only test Maize (C) since this is what the vast majority (~90%) of small and medium scale mills will consume.

3. **BATCH**: The third component will indicate Batch, numbered 1-10.

4. **SAMPLE**: The last component will indicate Sample, numbered 1-6.

An example label would be A – C – 3 – 1, meaning the first sample from the third batch of fortified Maize flour milled at mill A. All non-fortified flour samples will be labeled as batch "0" (e.g. A - C - 0 - 1). Whole grain samples will be labeled as Maize 1-6 only.

**Sample collection procedure:**

Each batch of grain is fully milled and the flour is collected in a bucket post mill. Once milling is complete, samples are taken from the bucket using a ladle (scoop). Touching the flour with human hands is avoided as much as possible.

Samples are to be taken from the following locations within the collection bucket:

- Sample 1: top surface of the flour near one side of the bucket
- Sample 2: top surface of the flour near the opposite side of the bucket
- Sample 3: three-quarters of the way down the bucket, near one side
- Sample 4: three-quarters of the way down the bucket, near the opposite side
- Sample 5: the middle of the bucket, after the flour has been mixed using the ladle
- Sample 6: the middle of the bucket, after the flour has been mixed again using the ladle