bioavid Diagnostics Procedure
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Standard procedure for verification of the performance of bioavid Lateral Flow Allergen Tests used **for new (unknown) solid samples.**

1 Find out, what dilution of the sample works for the Lateral Flow Stick.

- A beginner should first run water as a sample to see, how and when the control band appears on the stick.
- Add running buffer and water into a Reaction Vial, incubate 5 to 10 min as stated in the appropriate IFU, insert the stick. Note the time until the control band appears. No test band should be visible at the reading time stated in the appropriate IFU.
- Make a 1 in 10 extraction of the sample according to the standard procedure as shown in our short film. Be aware the fact, that some samples (e.g. chocolate bars) are not easy to homogenize – difficult samples may require more sophisticated instrumentation. Some fatty samples should be frozen in the blender vessel before crashing to avoid smear formation and to obtain a powdered sample.
- Run the supernatant of your extract in a Reaction Vial. If the control band appears only after one minute or even later, then dilute the sample extract 1 in 2 with water and run again. Repeat until the control band appears first time in less than a minute. Note the dilution required (= "working strength"; we haven't found a sample that required more than 1 in 100 dilution).
- The sensitivity of the test will be affected, if dilution is higher than 1 in 10. However, usually sensitivity of the test is high enough to find critical concentrations.
- If the sample was positive at the working strength of the sample, and fresh water was negative, the ingredients should be tested separately, starting again the procedure at 1. When negative, continue with step 2.

2 Determine the LOD for this sample.

- Make an 1 in 10 extract of your analyte (e.g. Hazelnuts) with saline applying the standard sample preparation procedure. This extract is defined as 100000 ppm of your analyte.
- Make a further 1 in 100 dilution to obtain a 1000 ppm concentration in saline or water, which will serve for spiking (e.g. 1 ml extract plus 99 ml water, mix thoroughly).
- Take 1 ml of the 1 in 10 extract of the sample prepared in step 1 and add 100 µl of the 1000 ppm spike to obtain a ca. 100 ppm concentration of the analyte in the sample.
- Dilute further as required to the working strength. Run the test according to the IFU. The result should be positive. The concentration compares to 1000 ppm in the sample before extraction.
- When positive, make a 100 ppm spike and add 100 µl of this spike into the sample extract of step 1. The concentration compares now to a 100 ppm contamination in the sample. Run again and find positive or negative.
- Repeat with higher or lower spike concentrations if more precise determination of LOD is required.

- This is a standard procedure for evaluation of the method for all new samples. It is laborious if executed first time. It is a routine for bioavid. Therefore, we offer this as a service for 90 Euros per sample and parameter. We just need to receive the sample. Usually we return the result within a week after receiving the sample.

Please contact us, if you have further questions.

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