

Product: OtaCLEAN SMART

According to the matrix please select the appropriate method.

- 1 For non fatty matrices recommended e.g. wheat, corn, rice:
 - Extract 20 g of sample with 100 mL (80 % methanol : 20 % water) in a blender jar at high speed for three minutes.
 - Pass the extract through a plaited filter.
 - Add 3 mL of the purified extract to 12 mL PBS buffer (pH 7.2).
 - Continue with 7.
 (Note: If there is a strong precipitation by mixing with PBS buffer, a further filter or centrifuge step is highly recommended before passing through the IAC column).

2 For fatty matrices recommended e.g. peanut, raisins, seed:

- Add 2.0 g NaCl to the sample.
- Extract with 100 mL of methanol : water (8:2) and 50 mL of n-hexane in a blender jar at high speed for three minutes.
- Pass the extract through a plaited filter. (Note: If there is a separation of phases to be found, the lower liquid phase is used for the following steps). To accelerate phase separation the extract could be centrifuged at 1000xg for 5 min.
- Add 3 mL of the purified extract to 12 mL PBS buffer (pH 7.2).
- Continue with 7.
- 3 For spices and animal feed e. g. black pepper, nutmeg, turmeric, chilli, pet food, animal feed:
 - Add 2.0 g NaCl to the sample.
 - Extract with 100 mL of methanol : water (8:2) and 50 mL of n-hexane in a blender jar at high speed for three minutes.
 - Pass the extract through a plaited filter. (Note: If there is a separation of phases to be found, the lower liquid phase is used for the following steps). To accelerate phase separation the extract could be centrifuged at 1000xg for 5 min.
 - Add 1 mL of the purified extract to 6 mL PBS buffer containing 8 % Tween20.
 - Continue with 7.

4 For coffee matrices recommended e.g. coffee beans, instant coffee:

- 5.0 g of the sample were homogenized and solved in 40 mL (50 % methanol / 50%, 3 % NaHCO3). 10 mL of this solution are mixed thoroughly with 10 mL of dichloromethane for 5 min. The upper liquid phase was mixed with 10 mL dichloromethane for 5 min.
- (Note: If there is a separation of phases to be found, the upper liquid phase is used for the following steps). To accelerate phase separation the extract could be centrifuged at 1000xg for 5 min.





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- Add 0.8 mL of the purified extract to 19.2 mL PBS buffer (pH 7.2).
- Continue with 7.

5 For wine matrices recommended e.g. red wine, white wine:

- 10 mL of the sample was mixed with 10 mL (1 % PEG, 5 % NaHCO3) for 3 min.
- The sample is passed through a plaited filter to remove precipitations.
- 3 mL of the sample was diluted with 12 mL PBS (pH 7.2).
- Continue with 7.

6 For beer matrices recommended:

- 20 mL of the sample were degassed by sonication.
- 8 mL of 3 % NaHCO3 solution was added and mixed well.
- The sample is passed through a plaited filter to remove precipitations and suspended particles.
- 3 mL of the sample was diluted with 12 mL PBS (pH 7.2).
- Continue with 7.

7 Immunoaffinity chromatography procedure:

- The diluted extract is passed through a 0.2 µ syringe filter.
- 1-10 mL (depending on the sensitivity of the detection) is applied on the OtaCLEAN SMART column. A gentle vacuum or overpressure may be used in all steps passing liquid through the column; nevertheless, it is indispensable to maintain a maximum flow rate of 1.5 mL/min.

(10 mL diluted extract (using protocol 1 or 2) represent 0.4 g matrix.

2.8 mL diluted extract (maximum applicable volume for this extraction using protocol 3) represent 0.08 g matrix.

10 mL diluted extract (using protocol 4) represent 0.05 g matrix.

10 mL diluted extract (using protocol 5) represent 1 g matrix).

10 mL diluted extract (using protocol 6) represent 1.428571 g matrix).

- To wash the column, pass 2 mL of distilled water through the column, which could be used to wash residual sample material from the reservoir.
- Carefully remove the residual water in the column.
- Elute with 0.4 mL of methanol; let the methanol act in the gel for 5 minutes.
- Dilute or concentrate eluate to your requirements and measure directly by HPLC with fluorescence detection. The use of a post-column derivatization is recommended!

If you have any questions, please contact: <u>mycotoxins@LCTech.de</u>





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