

## DNA extraction of fat containing samples using n-hexane and SureFood® PREP kits

This protocol is recommended for samples with a high content of fats or oils, such as **mayonnaise, spread, lard, lecithin, butter, cocoa butter (point 3 and 4)**. Please also refer to point 5 and Table 1 for additional matrices and deviating recommendations.

### 1. Sample preparation

Please prepare the samples in duplicates.

### 2. Additional required equipment and materials

- micro balance for weighing the sample
- 1.5 ml, 2.0ml and 5.0 ml reaction tubes (not supplied with the kits)
- n-hexane (purity  $\geq$  95%, not supplied with the kits)
- Vortex mixer
- heating block (up to 65°C)
- micro centrifuge (up to 12,000 rpm)

### 3. Procedure for fats or oils using SureFood® PREP Advanced protocol 1

- Transfer 500  $\mu$ L or 500 mg oil or fat into a 5.0 ml tube
- Add 1.0 ml n-hexane
- Mix briefly on a Vortex mixer
- Add 900  $\mu$ l Lysis Buffer and 20  $\mu$ l Proteinase K
- Mix briefly on a Vortex mixer
- Incubate on a heating block under continuously shaking for 60 min at 65°C
- Centrifuge for 2 min at 12,000 rpm
- The aqueous phase is on the bottom of the tube - transfer it with a pipette into a new tube (not supplied with the kit)
- Take the aqueous phase for further preparation
- Continue with step 3 Pre-filtration and setting of optimal binding conditions (without the first centrifugation step for 1 min at 12,000 rpm)

### 4. Procedure for fats or oils using SureFood® PREP Basic

- Transfer 200  $\mu$ L or 200 mg oil or fat into a 5.0 ml tube
- Add 1.0 ml n-hexane
- Mix briefly on a Vortex mixer
- Add 400  $\mu$ l Lysis Buffer and 20  $\mu$ l Proteinase K
- Mix briefly on a Vortex mixer
- Incubate on a heating block under continuously shaking for 30 min at 65°C
- Centrifuge for 2 min at 12,000 rpm
- The aqueous phase is on the bottom of the tube - transfer it with a pipette into a new tube (not supplied with the kit)
- Take the aqueous phase for further preparation
- Continue with step 3 Pre-filtration and setting of optimal binding conditions (without the first centrifugation step for 1 min at 12,000 rpm)
- Elution with 50  $\mu$ L

### 5. Procedure for additional matrices and deviating recommendations

- Transfer sample, according to User Manual or as indicated in table 1, into a 2.0 ml tube
- Add n-hexane as shown in table 1
- Mix briefly on a Vortex mixer
- Add 400  $\mu$ l or 580  $\mu$ l\* Lysis Buffer and 20  $\mu$ l Proteinase K
- Mix briefly on a Vortex mixer
- Incubate on a heating block under continuously shaking for 30 or 60 min\* at 65°C
- Centrifuge for 2 min at 12,000 rpm
- The aqueous phase is on the bottom of the tube - transfer it with a pipette into a new tube (not supplied with the kit)
- Take the aqueous phase for further preparation
- Continue with step 3 Pre-filtration and setting of optimal binding conditions, point 2.3 Extraction protocol from SureFood® PREP Advanced protocol 1 or SureFood® PREP Basic (without the first centrifugation step for 1 min at 12,000 rpm)

\*according to User Manual SureFood® PREP Advanced or SureFood® PREP Basic

## DNA extraction of fat containing samples using n-hexane and SureFood® PREP kits

February 2025

Table 1

matrices	application	Prep/protocol	devitating treatment
chips	Allergen	PREP Advanced Protocol 1	add 200 µl n-hexane
creme fraiche	Allergen	PREP Advanced Protocol 1	take 200 mg sample, add 200 µl n-hexane
dressing, French Dressing, Aioli	Allergen	PREP Advanced Protocol 1	if necessary setting the pH value, take 200 mg sample, add 300 µl n-hexane
pesto	Allergen	PREP Advanced Protocol 1	take 200 mg sample, add 300 µl n-hexane
pizza	Allergen	PREP Advanced Protocol 1	take 200 mg sample, add 300 µl n-hexane
sausage products (cooked sausage, meat paste, ham and more), white sausage, liver sausage	Allergen	PREP Advanced Protocol 1	add 300 µl n-hexane
cheese (cream cheese, hard cheese)	Allergen, Animal	PREP Advanced Protocol 1	take 200 mg sample, add 300 n-hexane
fat, oil, samples with high content of fat (mayonnaise, spread, lard, lecithin, butter, cocoa butter)	GMO, Plant	PREP Basic	Procedure for fats or oils using SureFood® PREP Basic
Salad with high content of fat, like mayonnaise (potato salad, meat salad)	Animal	PREP Basic	take 100 mg sample, add 300 µL n-hexane, elution with 50 µL