#

Study Proposal

Assessment of the effectiveness of an aversive agent
in soluble film for liquid laundry detergent capsules

## Background

Due to the occurrence of several ingestion incidents with liquid laundry detergent capsules, especially involving young children, the detergent industry has introduced precautionary measures through a voluntary Product Stewardship Programme. With a view to further reducing the number of these incidents, additional measures will be introduced, including the use of an aversive agent in the soluble film of the capsules. In addition, use of an aversive agent in the film is also being proposed as a mandatory measure under an amendment of the CLP legislation.

To demonstrate the effectiveness of an aversive agent in this application, and to define the appropriate level of the aversive agent in the film, taste testing, ideally with the target audience, is required. This study proposal defines objectives and methodology for such a test.

## Objectives

The objectives of this test are:

1. Development of a method for measuring the oral rejection time, as a function of the level of aversive agent present in water-soluble film of detergent capsules.
2. Proof of the concept (i.e. feasibility of the test method, as well as effectiveness of the aversive agent) with one specific commonly-used aversive agent (denatonium benzoate) and one specific film grade (polyvinyl alcohol).

## Hypothesis of the study

In case of ingestion incidents with water-soluble detergent capsules, children put these capsules into their mouth and keep them in for some time. Eventually the capsules leak or break, releasing the detergent content, leading to strong vomiting, coughing, and in a small number of cases aspiration of detergent and gastric fluids.

The addition of an aversive agent into the water-soluble film will ensure that children take the capsules out of their mouths within seconds once they recognize the bad taste. Consequently, the aversive agent in the film is expected to help reduce the number of oral exposures that lead to actual ingestion of the detergent content.

## General study description

The response of the test panellists (young adults) to tasting water-soluble film with different levels of aversive agent will be observed. From this, a dose-response relationship will be established that links the deterring effect (rejection of the film) with the level of the aversive agent. Ultimately, the study should allow determining an appropriate level that leads to rejection within a pre-defined safe timeframe.

The test panel shall consist of young adults, as a proxy for the target audience for the safety measures on liquid laundry detergent capsules (i.e. young children). There are reliable indications that for bitter taste, and specifically for the common bittering agent denatonium benzoate, the response of adults is at best similar to children, and usually children are more sensitive.

Because of variations in individual sensitivity, a limited number of panellists may be insensitive to the studied aversive agent. If the test results indicate absence of sensitivity in a panellist, and this is confirmed in a subsequent taste test, the results for this panellist shall be excluded from the study.

The test product is the water-soluble film treated with (different levels of) the aversive agent. The film should be used in isolation, wrapped around a neutral object such as a coffee stirrer, to be taken into the mouth for tasting. Actual detergent capsules should not be used, to ensure the safety of the panellists.

Each panellist will be given a spatula with soluble film treated with a given level of aversive agent, and be asked to put and keep this into the mouth. It will then be recorded whether the panellist spontaneously rejects the spatula and if so, how long it was kept into the mouth prior to rejection. Panellists are only allowed to participate once. Otherwise, their prior experience with a bad tasting spatula would bias their behaviour and might lead to faster rejection times, at least partly driven by psychological factors instead of entirely being an actual response to the aversive agent.

A concentration series should be tested using this approach. First, in a screening round, a broad range of levels between 0 and (up to) 10000 ppm of aversive agent in film should be assessed. Next, based on the screening results, suitable aversive agent levels should be defined for a definitive testing round, aiming to refine the dose-response relationship for those levels leading to rejection between 0 and 10 seconds. As such, the study should develop a dose-response relationship between the level of aversive agent in the film and the average time it takes for rejection. This refined curve can then be used to determine the level that leads to rejection within 5 seconds. Further, lower and upper threshold levels may be established, respectively below which rejection does not occur, and above which rejection time does not decrease.

Note that for all test levels to be included in the study, absence of toxicological risk should be demonstrated in a risk assessment.

## Test material

The test material is a combination of one specific water-soluble film type with one specific aversive agent, at different concentration levels. Both the water-soluble film and the aversive agent tested shall be identified in the study report and/or in the study sponsor’s confidential study placement documentation. The results of the study are specific to the type/grade of water-soluble film and the type/grade of aversive agent used. Consequently, results cannot be extrapolated to substantially different combinations of film and aversive agent.

### Preparation of water-soluble film treated with aversive agent

Water-soluble films with different levels of the aversive agent shall be prepared:

* Screening test: untreated (blank) - 10ppm - 100ppm - 1000ppm - 10000ppm (\*)

(\*) if toxicological concerns exist with the higher screening levels, an alternative concentration series with lower levels should be used

* Final test: untreated (blank) + 4 levels to be determined based on the outcome of the screening test

Accuracy of the aversive agent’s levels in the film, and homogeneity of its distribution, shall be ensured by the producer of the treated film.

### Preparation of spatulas for taste testing

The treated water-soluble films shall be cut into strips of 3cm by 5cm, and shall be wrapped around standard wooden medical spatulas. Each strip shall be applied at one end of the spatula. To ensure adhesion, the contact area of the film to the spatula shall be slightly moistened before it is applied. Next, the film strip shall be tightly wound around the spatula. The end of the strip shall again be slightly moistened and pressed against the film already around the spatula, so it stays in place.

The spatulas with treated film shall be individually labelled using a coding system that does not disclose the level of aversive agent, neither to the panellists nor to the persons directly handing the spatulas to the panellists (double-blind approach).

Preparing the spatulas as well as labelling them, shall be done by the producer of the treated film.

### Specific - test material for the proof of concept

* Film: Monosol M8630. According to the patent literature Monosol’s M8630 water-soluble film is a commonly used grade for detergent capsules.
* Aversive agent: Bitrex. The bittering agent denatonium benzoate is widely used and state-of-the art. One supplier is marketing this under the trade name Bitrex, but it is available from different suppliers with the identical chemical active substance.

## Test Panel

### Panel composition

A test panel with 100 participants is required to conduct this study for one film / aversive agent combination.

The test panel shall consist of the following individuals:

young adults, in the age group of 18-25 years old

* equal mix male / female
* exclusion criteria:
	+ panellists with prior experience on tests of aversive agents should be excluded
	+ results from panellists who, during the test, are shown to be non-sensitive to the aversive agent being tested, shall be excluded from further data processing.

Each panellist shall participate to only one single tasting session, to avoid a biased response driven by prior experience.

## Test Design and Instructions

The test shall be conducted in two rounds:

1. a screening round in which a wide range of levels of the aversive agent is assessed;
2. a final round in which the dose-response relation is refined.

In both rounds, there shall be 4 test concentrations in addition to a blank (untreated film), with 10 replicates for each concentration. Hence, in total, there will be 100 tasting sessions (5x10=50 for the screening round, and 5x10=50 for the final round). TO NOTE: NUMBER OF CONCENTRATIONS PER ROUND, THE NEED FOR A BLANK DURING THE FINAL ROUND, AND THE NUMBER OF REPLICATES PER CONCENTRATION LEVEL, ARE INITIAL PROPOSALS ONLY AND MAY BE MODIFIED BASED ON GUIDANCE FROM THE LAB THAT WILL EXECUTE THE STUDY

The levels of aversive agent for the screening round are predetermined. The levels for the final round are to be defined based on the screening results. A recommendation shall be provided to the study sponsor within maximum one week upon finalisation of the screening round. The final round can only be organized several weeks after the screening round, to allow for preparation and shipment of the film and preparation of the spatulas.

For every tasting session (one panellist, one level of the aversive agent):

1. The test shall be conducted such that participating panellists cannot see the reaction of others in the test, and cannot talk to others who have just completed the test. Neither the panellists nor the persons providing the test samples to the panellists shall be informed of the presence and/or level of the aversive agent in the sample.
2. The panellist shall drink a defined small amount (50ml) of still water.
3. The panellist shall put the end of the spatula with the film into the mouth, ensuring contact with the tongue, and keep it there. No further instructions are to be provided.
4. Start the chronometer and allow 60 seconds to pass. Do not provide instructions to the panellist.
5. If the panellist has not spontaneously rejected the spatula after 60 seconds, allow the panellist to remove the spatula. To note: it may be that this panellist is non-sensitive to the aversive agent, which will require separate follow-up - see below.
6. Record whether the panellist has rejected the spatula within 60 seconds, and if so, after how many seconds the rejection occurred.
7. Provide a piece of dark chocolate to the panellist as a remedy to get rid of the unpleasant taste. Also provide a glass of water.
8. Exclude the panellist from any further participation to this test or similar tests in the future.

### Exclusion of non-sensitive panellists

Upon the processing of the data, it may be found that some panellists did not reject the spatula, whereas at least 75% of the replicates for this aversive agent level have led to rejection. This may be due to individual non-sensitivity, which is an exclusion criterion for the study. Additional follow-up with these panellists is needed to assess whether they are indeed non-sensitive. This is expected to be needed only for a very low percentage of the panellists.

Assessment of sensitivity to the aversive agent under study is to be done by observing the panellist’s reaction to drinking orange juice containing a pre-defined level of aversive agent that is known to lead to rejection (\*).

1. The panellist should not be made aware of the objective of this follow-up test.
2. Let the panellist drink a small amount (50ml) of normal orange juice.
3. Let the panellist drink a glass (200ml) of orange juice treated with the aversive agent. Do not inform the panellist that the taste might be bad.
4. If the panellist does not spit out the treated juice while drinking it, and drinks more than half of the glass (>100ml), the panellist’s results shall be excluded from the study.
5. Subsequently, the panellist shall be given a piece of dark chocolate as a remedy to get rid of the unpleasant taste. Also a glass of water shall be provided.

 (\*) for the proof of concept with Bitrex, a level of 10ppm should be used - cf. Sibert & Frude, Arch. Emerg. Med. 8:1-7 (1991)

## Reporting of results

All raw data collected during the study shall be reported, except the identities of the panellists, that are to remain confidential to the testing laboratory. Note that these identities shall be archived by the testing laboratory for further reference to avoid their participation in other similar studies in the future.

The ultimate deliverable of the study is a dose-response relationship, expressing the average rejection time among panellists as a function of the level of the aversive agent in the film. This dose-response relationship shall be presented as a curve as well as a table. From this, it shall be determined and reported what is the aversive agent concentration that is expected to lead to rejection, on average, within 5 seconds. The 95% confidence interval for this value shall also be provided.

In addition, based on the screening round, an indication shall be provided of the aversive agent level below which no rejection is expected; as well as the level above which no further reduction time is anticipated.

## Study Limitations

The test design will be able to demonstrate the effectiveness of an aversive agent in a virgin water-soluble film, and to determine the level of aversive agent required to achieve rejection within a critical time frame. Individual detergent producers will be responsible to ensure that this required level be present in the film under actual use conditions, i.e. for detergent filled capsules also after storage (taking into account the possible migration of the aversive agent from the film into the detergent). Such assessment is not within the scope of this study.