DERMAL IRRITATION / CORROSION Trial	
Our reference: **1419314**	Your ref.**: BOLDFOAM F-40 BATCH NR F40021101**
Sample description: Liquid, pale yellow in colour and transparent	
Conservation: In darkness and refrigeration (5±3ºC) max. 2 months	
Sample preparation: Not required	
Trial start date: 17/05/2011	Trial end date: 20/05/2011

Dermal irritation manifests as reversible inflammatory modifications to the skin, following the application of the test substance for an exposure time of 4 hours. Corrosion manifests as irreversible damage to the skin following a maximum exposure time of 4 hours.

The trial is extended over an observation period of 72 hours, however, observation should last for up to 14 days, when the reversibility of lesions must be established.

The trial method is based on the current applicable regulation in this field:

- Method B.4 which comes under Regulation (EC) No. 440/2008 equivalent to OECD method 404 (2002).

It is a liquid sample, with a pH range falling between 2.0 and 11.5, which is applied to the skin of two animals for a period of 4 hours. Depending on the results observed in the application area, the trial will be continued on a third animal, or perhaps it will not be necessary to test the sample on more animals.

MATERIAL AND METHOD

SELECTED SPECIES

Young and healthy albino rabbits are used in this trial, with a body-weight over 1.750 Kg. The selected breed is NEW ZEALAND WHITE. The animals are supplied by our accredited provider, CHARLES RIVER LABORATORIES (France), at the approximate age of 8 weeks. The breeding records and health verifications of the animals are stored in our animal facilities files.

Living quarters and maintenance

Upon arrival to the laboratory, the animals are inspected by an expert technician, who assesses their state of health, then they are placed in correctly labelled individual stainless steel cages.

The animals are acclimatised to their living quarters for a minimum period of 20 days. They are observed daily to detect the possible appearance of any type of pathology or behavioural changes. When the acclimatisation period is complete, the animals are assigned to each trial group. The environmental conditions are checked every day, remaining within the following intervals:

- Temperature at 20 ℃ ± 3 ℃
- Relative humidity rate between 30 and 70%
- Photoperiod of 12/12 hours of light/darkness

The animals have *ad libitum* access to food and water throughout the acclimatisation and trial period. The diet is granulated feed designed for rabbits, supplied by our authorised provider, Harlan Ibérica, S.L.

Experimentation groups

The distribution of sexes is not relevant. If females are used, they should be nulliparous and not pregnant. In each animal, the surfaces next to the treated skin constitute the negative control area. The animals have a trial starting age of 12 weeks, with an initial weight of 2.6 Kg and end weight of 2.7 Kg.

SAMPLE PREPARATION

The sample, in liquid form, is applied directly to the skin, with no previous treatment required. The exposure will be carried out with a single dosage level (0.5 g / animal).

OPERATIONAL MODE

The application zone, on the dorsal region of the trunk of the animals, is carefully shorn before exposure, with the aim of not damaging the skin and to ensure close contact with the trial substance, over a minimum surface area of 6cm². The sample is applied to this zone, covered with a self-adhesive, hypoallergenic and semi-occlusive dressing (Mepore® or similar), secured firmly around the torso of the animal with a cotton bandage and a tubular elastic bandage during the contact time. Once the exposure time is complete, the dressings are removed and the trial substance is washed off with water.

The contact time is 4 hours. If the reaction is conclusive in the first two animals, further animals will not be required for exposure. If the reaction observed is inconclusive, the trial should be performed on a third animal. To assess ambiguous reactions, exposure on more animals may be necessary.

OBSERVATIONS

Given that no reaction was shown on the skin of either of the two animals, no further animals are required for testing.

The assessment of the reactions is carried out at 24, 48 and 72 hours following exposure. With the aim of establishing the reversibility of the lesions, the observation period is extended until the effects have disappeared completely, with a maximum duration of 14 days.

RESULTS

The results given in this report have been assessed in accordance with the following value table, depending on the criteria stipulated in the current regulations:

ASSESSMENT TABLE

FORMATION OF ERYTHEMA AND SORES	VALUE
- No erythema	0
- Very slight erythema (barely visible)	1
- Clearly defined erythema	2
- Moderate to serious erythema	3
- Serious erythema (red-violet colour) with ulceration or sores (deep lesion)	4

FORMATION OF OEDEMA	VALUE
- No oedema	0
- Very slight oedema (barely visible)	1
- Light oedema (outside of the area is clearly outlined by a clean raised section)	2
- Moderate oedema (approx. elevation 1mm)	3
- Serious oedema (inflammation of over 1mm which extends over the exposed area)	4

RESULTS TABLE

ASSESSMENT OF CUTANEOUS LESIONS

FORMATION OF ERYTHEMA AND SORES

ANIMAL No.	24 h.	48 h.	72 h.	AVERAGE
1	0	0	0	**0.00**
2	0	0	0	**0.00**

FORMATION OF OEDEMA

ANIMAL No.	24 h.	48 h.	72 h.	AVERAGE
1	0	0	0	**0.00**
2	0	0	0	**0.00**

OVERALL ASSESSMENT

	ERYTHEMA	OEDEMA
No. of animals with persisting lesions at the end of the trial	**0 / 2**	**0 / 2**
No. of animals with average value ≥ ε 2,3	**0 / 2**	**0 / 2**

CONCLUSIONS

As indicated in the results table, no erythema or oedema were observed in the treated area of any of the animals used in the experiment.

According to Regulation (EC) 1272/2008 governing the classification, packaging and labelling of substances and mixtures, any substance whose average value produces erythema or oedema equalling 2.3 or above in two or more animals, is considered to be an irritant, or whenever damage persists to the end of the observation period of 14 days in at least two animals. It is considered to be corrosive when irreversible damage is observed on the exposed area of skin.

To conclude, we can affirm that in the conditions of this trial, the sample is from a toxicological perspective and can be legally classified as follows:

NON-IRRITANT – Non-assignable to any cutaneous irritation or corrosion category