



**Trial Report
n° M9/2006**

**lactium® stability and
activity in Fruit Juice**



TRIALS AIM :

- To observe the impact of lactium[®] on the quality attributes of a ***Fruit Juice*** (aspect, flavour & taste).

- To quantify lactium[®] in the finished product.

- To validate lactium[®] activity in a ***Fruit Juice*** after different shelf life times :
 - fresh product,
 - 7 months shelf life.

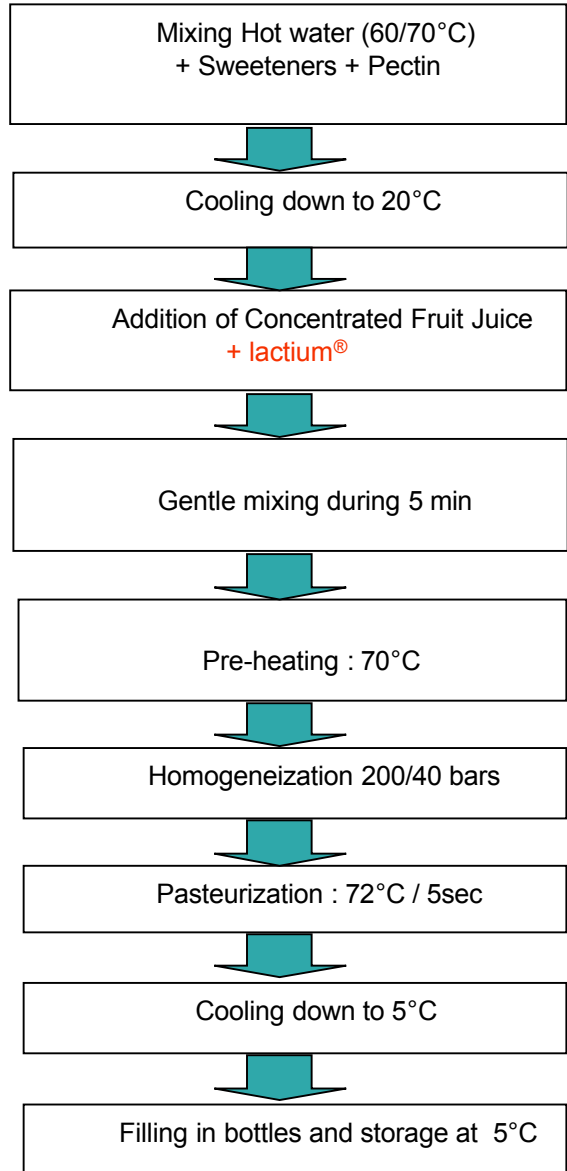


Fruit Juice

1- MATERIAL & METHODS

➤ Fruit Juice Recipe & Process

Raw Materials	Fruit Juice	Fruit Juice with lactium®
Water	85.00%	85.00%
Concentrated Fruit Juice (>70%)	14.60%	14.60%
Sweetener	0.10%	0.10%
Stabiliser (HM-Pectin) + citric acid	0.30%	0.30%
lactium®		+ 0.6 g/L
Total	100.00%	100.00%



➤ **Visual appearance :**

minor difference between the Fruit Juice with lactium® (more whitish) and the reference

➤ **Physical stability :**

No separation neither sedimentation after 5 weeks at 7°C for the 2 products

➤ **Sensory properties (triangle test) :**

no significant difference between the Fruit Juice with lactium® and the reference

➤ **Recovery of α_{s1} -CN-(91-100) peptide :**

92% after 5 weeks at 7°C compared to the untreated product



1- MATERIAL & METHODS

➤ Conditioned Defensive Burying method

The Relaxing effect of Fruit Juice containing lactium® was tested on Conditioned Defensive Burying method at different times :

- fresh Fruit Juice,
- Fruit Juice after 7 months of shelf life.

Fruit Juice had been orally administered to the male Wistar rat :

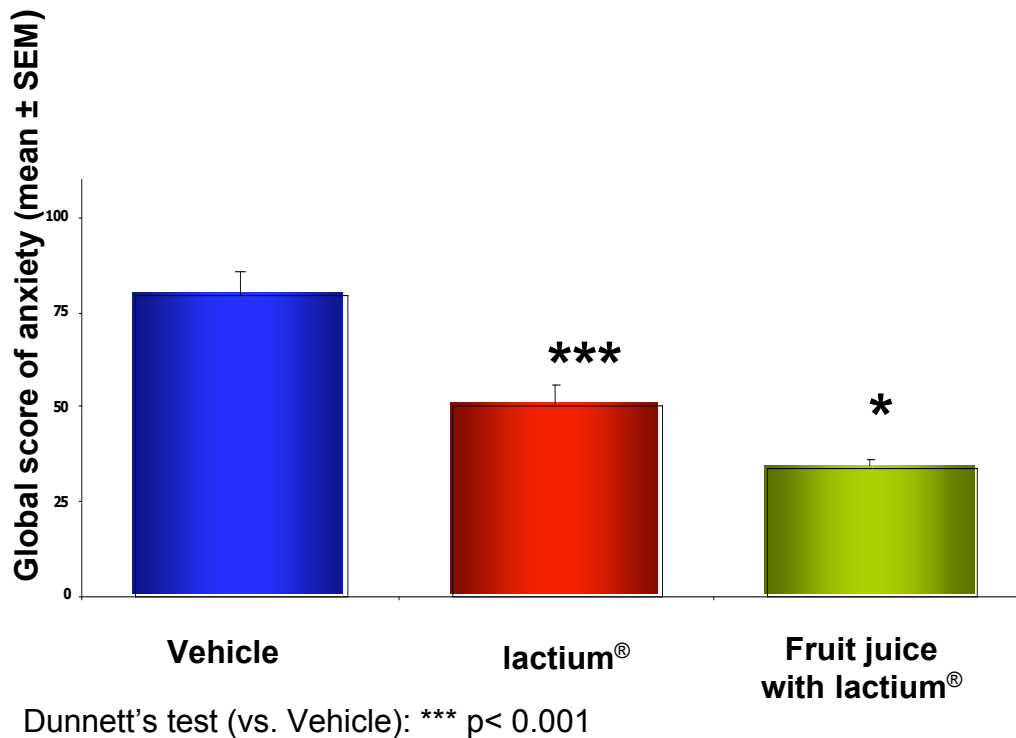
- Acclimatization of the rats during a period of 7 days
- 3 groups of 12 rats:
 - vehicle,
 - lactium® (reference),
 - fresh Fruit Juice with lactium®.
- 4 groups of 12 rats:
 - vehicle,
 - lactium® (reference),
 - 7-months Fruit Juice without lactium® ,
 - 7-months Fruit Juice with lactium®.
- Fruit Juices were lyophilized after proper ageing.
- Fruit Juice were administered so that rats ingested 15 mg/kg of lactium® which corresponds to the active dose in rats
- Oral administration was made 1 hour before the test
- After shock administration (stressing event), the behavior of the rats were recorded during 5 minutes
- Global anxiety score = Sum of the rankings for three parameters:
 - duration of probe-burying;
 - number of head stretching towards the probe
 - percentage of approaches towards the probe followed by retreats



Fruit Juice
2- RESULTS

➤ Fresh Fruit Juice

Products	Vehicle (n = 12)	lactium® (n = 12)	Fruit juice with lactium® (n = 12)
Mean ± SEM	79.55 ± 6.54	50.46 ± 5.62	34.13 ± 1.56
ANOVA F(3.43)=14.68; P<0.0001 Dunnett's t test (vs. Vehicle) Significance		t=3.39 P<0.005	t=7.04 P<0.001

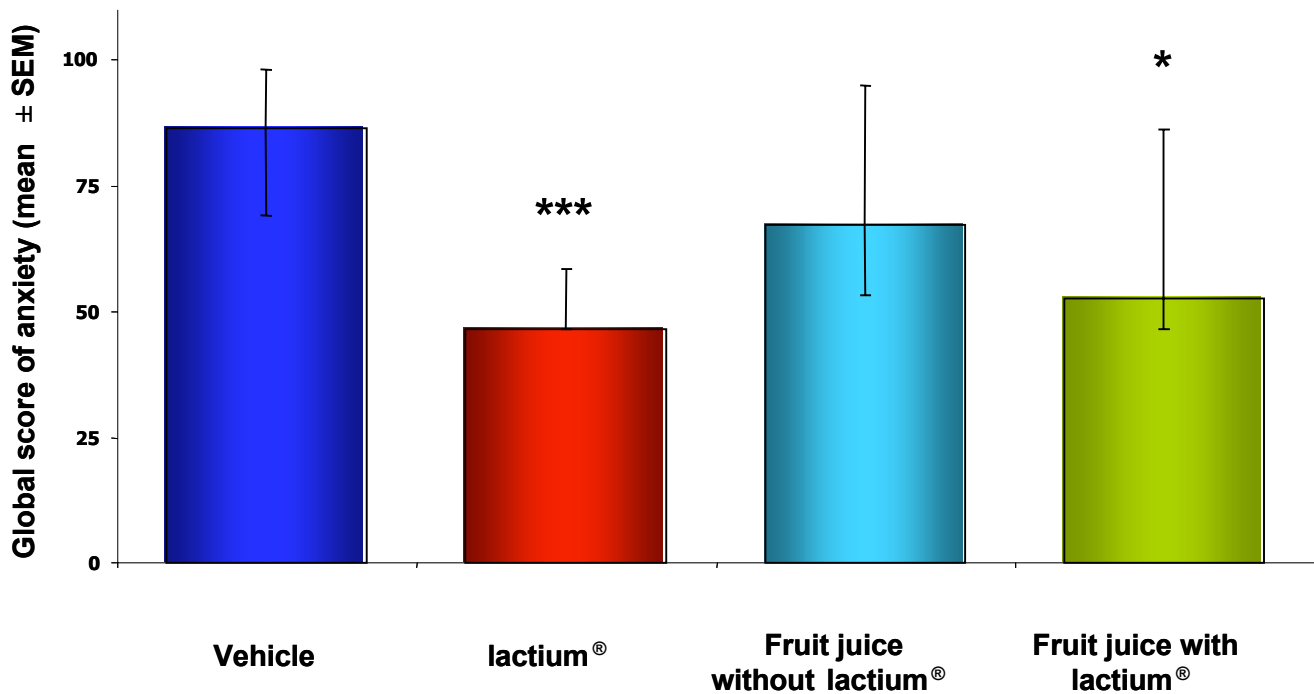




Fruit Juice
2- RESULTS

➤ 7-months Fruit Juice

Products	Vehicle (n = 12)	lactium® (n = 12)	Fruit juice with lactium® (n = 12)	Fruit juice without lactium® (n = 12)
Mean	86.50	46.50	67.25	52.52
Qi - Qs	69 - 98	46.5 – 58.25	53.25 – 94.75	46.5 - 86
Kruskal-Wallis test H(df=3)=11.41; P<0.01 Mann-Whitney U-test (vs. Vehicle) Significance		U=18.00 P<0.005	U=48.5 NS	U=34.00 P<0.05



Mann-Whitney U-test (vs. Vehicle) : *p<0.05; ***p<0.005



Fruit Juice

3- COMMENTS

- The addition of lactium® has a minor impact on the visual appearance of the fruit juice. No sedimentation is observed after 5 weeks and no sensory difference has been detected.
- The recovery of lactium® in the finished product with an HPLC method (#MLB C119B) is almost total.
- Fresh and 7-months shelf-life Fruit Juice containing lactium®, orally administered, significantly decrease the global score of anxiety of rats submitted to a conditioned defensive burying test, in comparison with vehicle and pure lactium®.
- It is not the case for Fruit Juice without lactium®.
- The thermal process and acidic pH do not denature lactium®.



CONCLUSIONS :

- ✓ The addition of lactium[®] has no significant impact on the quality attributes of a Fruit Juice (aspect, flavour & taste)
- ✓ The Fruit Juice process does not decrease the activity of lactium[®].
- ✓ Moreover, after 7-months of shelf-life, the Fruit Juice with lactium[®] is still active and reduces significantly the symptom of stress.
- ✓ lactium[®] is almost totally recovered in the finished product.