MOLECULAR ACTIVATION

This book compiles renowned works related to the study and application of the most effective antioxidants in all types of degenerative diseases.

The molecular activation process is demonstrated by various methods of analysis which explain the difference between the antioxidants used and their activated molecules, and factors which have an impact on the activation mechanism are described.

Different types of natural antioxidants (inorganic, organic, biological and biochemical) are studied and compared; some of these are products of the company, ***CATALYSIS, S.L****.*

The molecular activation process is demonstrated by various methods of analysis which explain the difference between the antioxidants used and their activated molecules, and factors which have an impact on the activation mechanism are described.

Molecular Activation

The molecular activation procedure considerably improves the biological activity and biochemical reactivity of all antioxidant molecules.

This method of activation is much more effective when applied to a far wider range of hydrosoluble and liposoluble molecules, and also when improving certain parameters of the procedure. It was discovered in the **C.S.I.C. (Consejo Superior de Investigaciones Científicas)**, Spain's most important public research body.

We know the answer to this activation in numerous antioxidants, but we still do not know the electron assimilation and accumulation mechanism which causes such a considerable increase in antioxidant capacity; neither do we know the mechanism by which accumulated electrons are able to reduce the free radicals in oxidant molecules.

In this mechanism we have observed greater synergy between some antioxidants used, that are sometimes capable of considerably increasing their overall antioxidant capacity.

Many factors can influence the activation of all antioxidants.

Amongst the most important chemical factors are: the molecular structure, the active functional groups, specific antioxidant catalysts, molecular weight, the pH, double carbon bonds, their coefficient of solubility, etc., as well as the antioxidant capacity of each molecule.

The duration and intensity of molecular activation are amongst the most influential **physical factors**.

Not all antioxidants require the same activation time to reach their maximum antioxidant capacity; the most important parameter for the control of better performance is their optimisation. Once their highest antioxidant capacity is at its most favourable peak, activation must be suspended because, after that maximum peak, their antioxidant capacity starts to diminish gradually or quickly. This zone - just after the optimal time of activation - is when oxidation begins and it is therefore necessary to fix the most appropriate time with great precision in each specific case.

When there is a mixture of two or more antioxidants, the optimal activation time is previously calculated for each preparation and this fixed parameter is always respected.

The antioxidant capacity is analysed by using the Somogyi-Nelson reagent for determining reducer substances; this reagent is capable of evaluating with mathematical precision the exact quantity of cupric ions which are reduced to cuprous ions. A fixed concentration of 1 mg/ml of antioxidant, or a mixture of the antioxidants that we want to assess, is used to homogenise these analytical calculations.

When some antioxidants, such as certain natural flavonoids, are activated, they can increase their antioxidant capacity by 618.64% (7.186 times higher than without activation); this is because synergetic effects also increase with certain antioxidants.

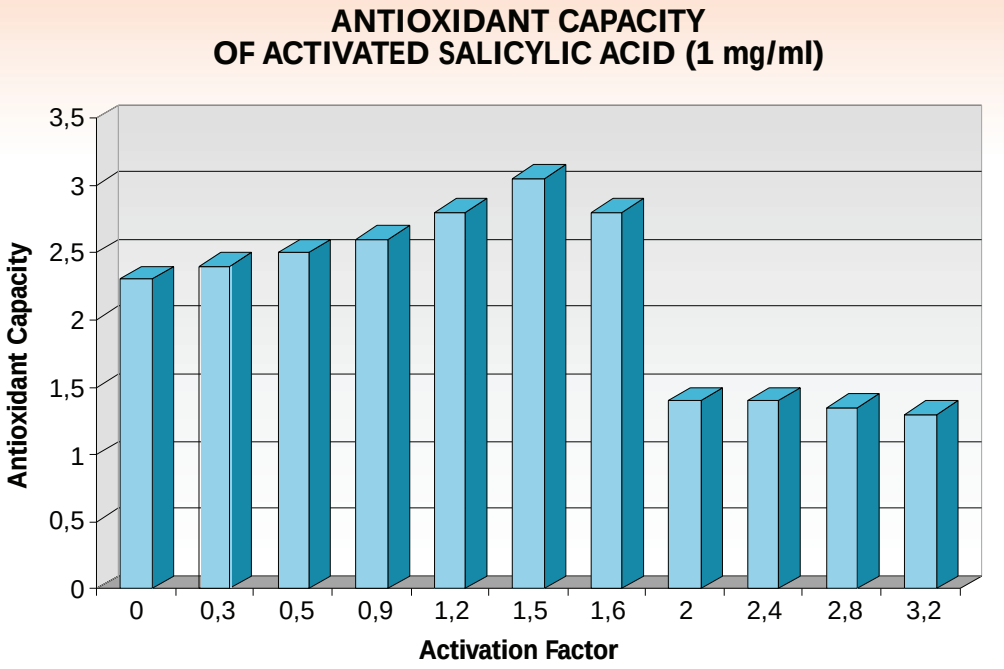
Sometimes, certain specific catalytic complexes can also favour activation reactions.

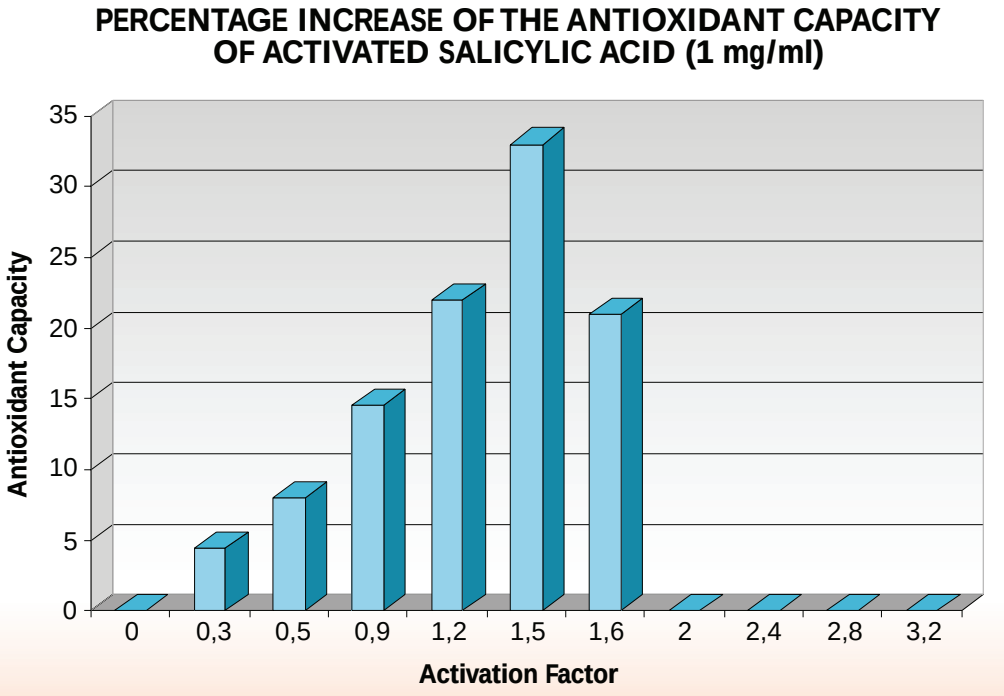
For a better understanding of the **importance of activation** we could give the example of liquid **VIUSID**.

In an inactivated state, it originally had an antioxidant capacity of 1.100 points; when activated, it rose to the following values: 1.225, 1.320 and 3.300, increasing by 200%. Later, in 2004, we managed to obtain an antioxidant capacity of 9.600 in liquid **VIUSID** (7.836 times higher than its original non-activated state).

These results demonstrate that **activation is indispensable and essential for obtaining the greatest biological activity and, as a consequence, the greatest effectiveness, in the treatment of diseases which directly or indirectly produce free radicals.**

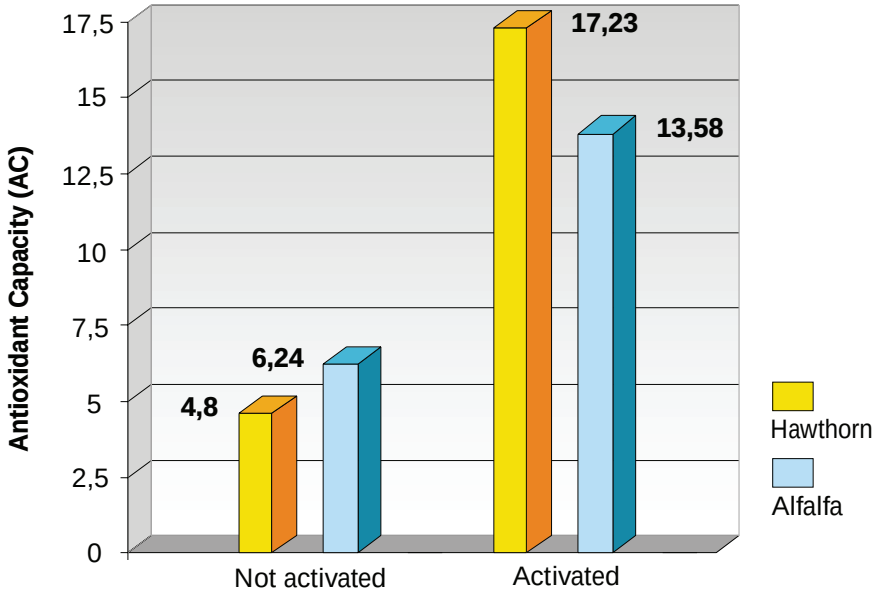
- *Antioxidant Capacity of Salicylic Acid*



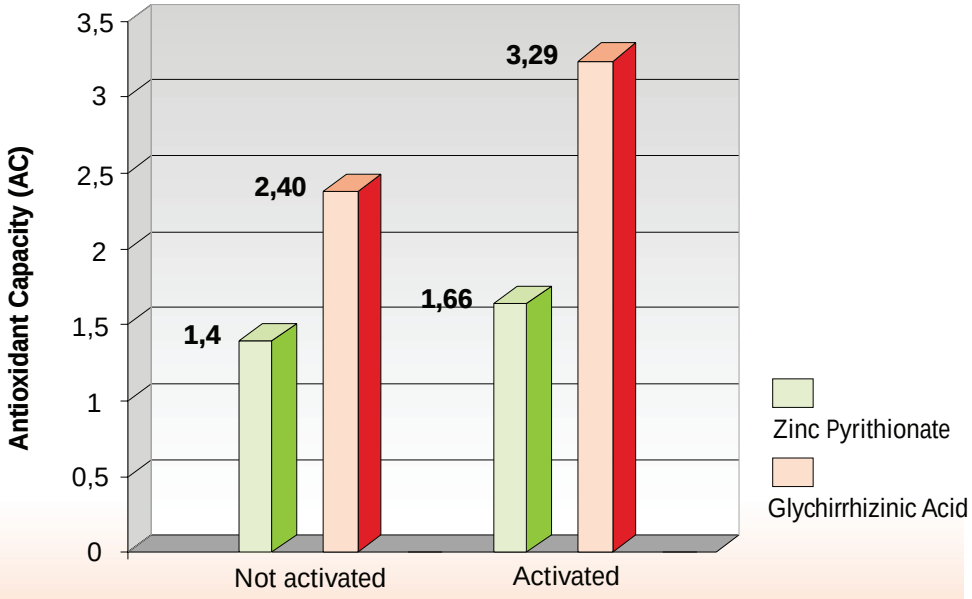


- ***Impact of Molecular Activation***
  on the Antioxidant Capacity of Espino Albar (Hawthorn) and Alfalfa Extracts, Zinc Pyrithionate and Glycirrhizinic Acid

IMPACT OF MOLECULAR ACTIVATION ON THE ANTIOXIDANT CAPACITY (AC) OF ESPINO ALBAR (HAWTHORN) AND ALFALFA EXTRACTS (Increase in AC)



IMPACT OF MOLECULAR ACTIVATION ON THE ANTIOXIDANT CAPACITY (AC) OF ZINC PYRITHIONATE AND GLYCIRRHIZINIC ACID (Increase in AC)



• *Biophysical Analyses* - *Ultraviolet*

The molecular activation procedure considerably improves the antioxidant capacity and the biological activity of activated antioxidants.

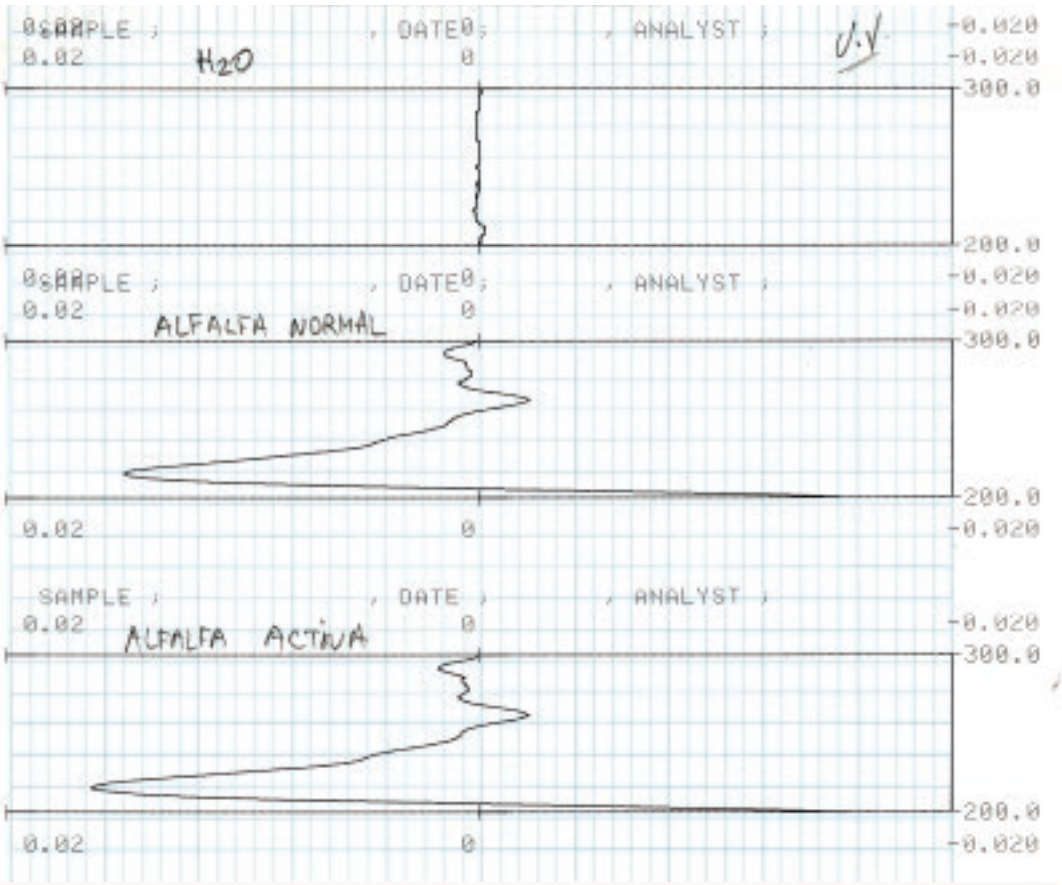
The activation time required to optimise the antioxidant capacity of a molecule varies between a few seconds and several hours, depending on the structure of each compound.

Results are more relevant in some molecules than in others, but it is an irreversible process and in all the compounds treated **there is a thermostable increase in the antioxidant capacity, which is not lost with time**.

Analyses made to determine possible structural changes between activated and non-activated alfalfa extracts lead to the following conclusions:

- The UV spectra of both extracts are very similar.
- There are no peaks.
- A differential spectrum is observed at 190 nm, which shows an increase in light absorption after the product has been activated.

UV Spectroscopy Studies



• *Biophysical Analyses* - *Ultraviolet*

The image reveals that the UV spectra are very similar in the extract of normal alfalfa and in activated alfalfa. Peaks neither appear, nor disappear. The apparatus was compensated with respect to the cuvettes and the base line was digitally removed and can only be seen on the X axis of the left-hand graph. That greater intensity close to 190 nm may indicate greater concentration of chromophoric or auxochromic groups, or higher condensation between groups of molecules, or an increase in aromaticity or resonant structures.

When the second derivative is obtained from the spectrum, valleys appear (where there had previously been peaks), marking not only the position and intensity of the highest points with precision, but also of the inflections. It is confirmed that no new peaks appear, but that light is better absorbed after activation. Differences are minimal. The apparatus is readjusted and it was proved that the second derivative from water (first graph on the left) is straight. The scale is considerably amplified. All the spectra (at that concentration of 0.1 mg/mL) appear in a range of 0.04 absorption units.

When the differential spectra are done (which is what was intended from the beginning), the explanation given above is visible, although more exaggerated. The base line is again accumulated and the two cuvettes are full of the same product (activated) in the left-hand graph. A straight line ought to be visible (see the range of the DO axis), because the same solution is being used as a target. Slight noise can be heard.

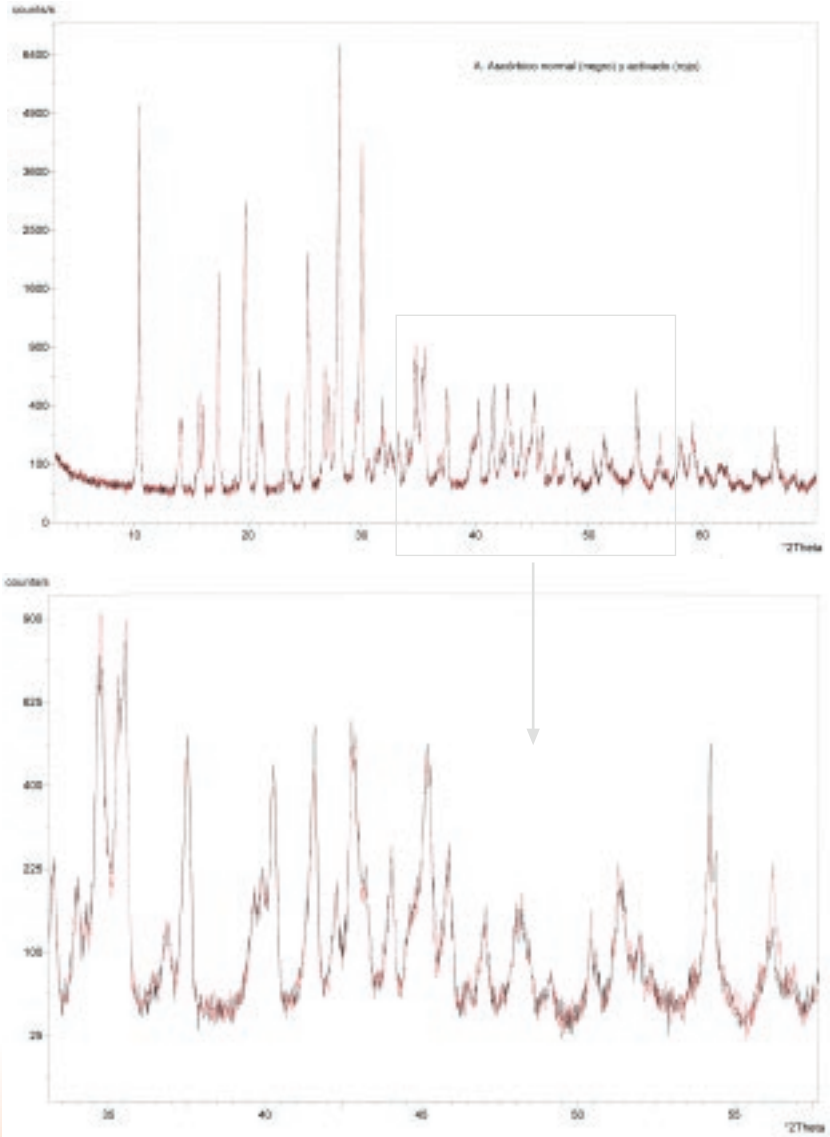
When activated and non-activated products were placed opposite each other in the respective exercises (reference and sample), a straight line or an equivalent noise would have appeared if there had been no changes in the product. A differential spectrum emerges which shows the wave longitudes where the light absorption of the product has increased after activation.

In this entire process, no obvious qualitative changes are observed.

Studies of X-rays (rXs)

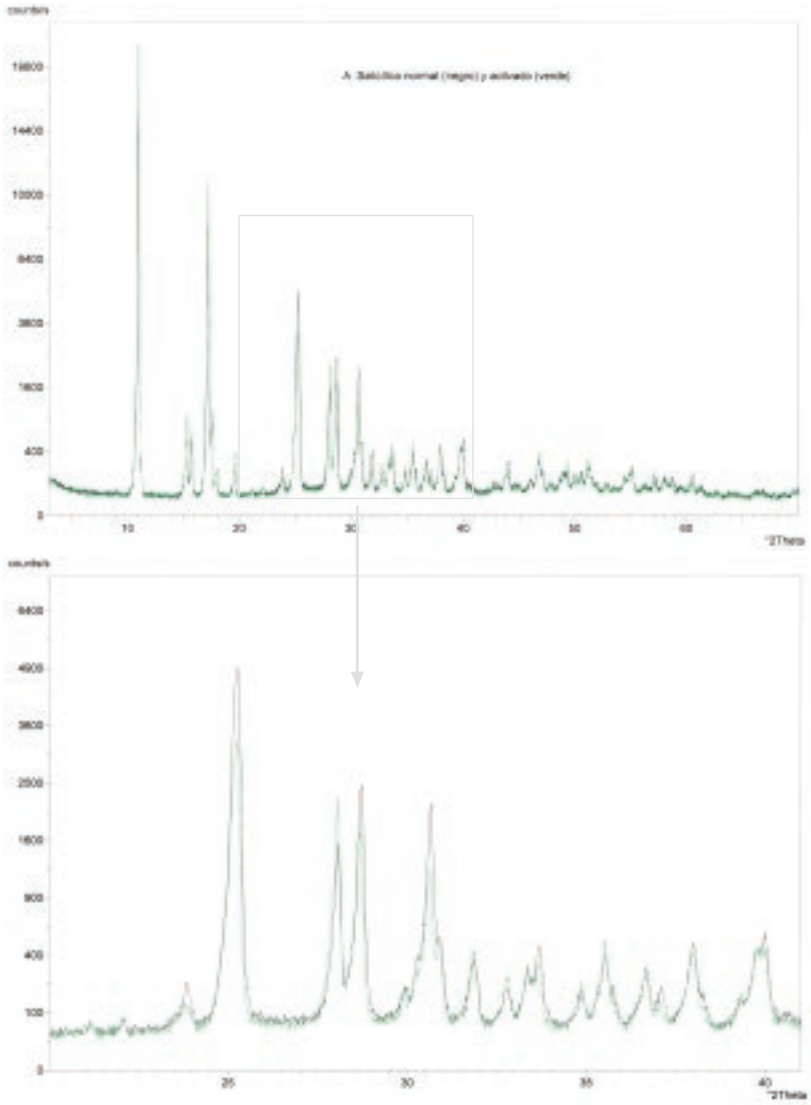
These are X-ray spectra of normal, activated ascorbic and salicylic acid. They are superimposed in pairs and different in colour. Diffractograms 2 and 4 are enlargements of the area where there seemed to be more differences; in any case, the differences are small and the spectroscopists comment that they are similar to what they expected when repeating the same sample with the same apparatus, although spreading the sample in a different way on the sample-bearer.

NORMAL VS. ACTIVATED ASCORBIC ACID
(Area with most changes)



- ***Biophysical Analyses*** *- X-rays*

NORMAL VS. ACTIVATED SALICYLIC ACID
(Area with most changes)



The differential analyses using X-ray techniques in two antioxidant molecules, such as activated and non-activated ascorbic and salicylic acid, lead to the following conclusions:

- There are no differences between these compounds, whether activated or not activated. The spectrum shows the same peaks at the same place.
- However, some of the peaks of the activated molecules of ascorbic and salicylic acid have a 25% higher intensity than the peaks of the non-activated molecules.

• *Biophysical Analyses* - *Electrical conductivity and Thermostability*

Electrical Conductivity Study of Extracts of Activated and Non-activated Alfalfa

- Fixed potential: 28V
- Intensity: CONTROL 150 mA
  ACTIVATED 172 mA

Percentage increase = 14,666 (greater polarity)

This increase of approximately 15% in the polarity of activated alfalfa indicates that the molecule has a higher electrical density, which means that, with greater polarity, the molecule can transport electrons more easily.

Thermostability Study of Extracts of Activated and Non-activated Alfalfa

After 10 minutes' boiling and 1 minute in a microwave:

- Loss of antioxidant capacity (AC): CONTROL - 1,20%
  ACTIVATED - 8,63% *****

The 7.43% difference implies greater reactivity power in the activated extract.

Thermostability Study of Activated and Non-activated Cysteine

After 10 minutes' boiling and 1 minute in a microwave:

- Both forms remained stable, without losing their antioxidant capacity (AC).

Thermostability Study of Activated and Non-activated VIUSID

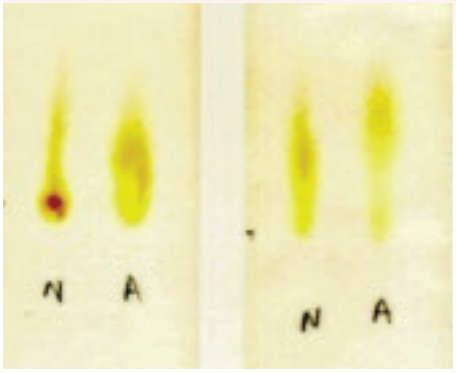
After 10 minutes' boiling and 1 minute in a microwave:

- Loss in the antioxidant capacity (AC) after boiling for 10 minutes is 2.47 times higher in activated **VIUSID** because its molecules possess greater reactivity power, due to possible glycosylation interaction and to the glucosamine in the product.
- Loss of antioxidant capacity (AC): CONTROL - 11,38%
  ACTIVATED - 28,13% *****

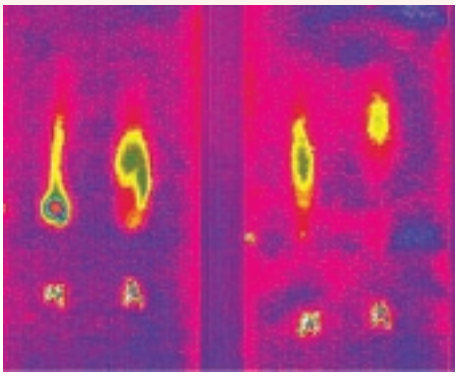
***** The higher loss caused by glycosylation involves greater reactivity between antioxidant molecules and the sugars in the medium.

• *Biophysical Analyses* - *Electrophoresis*

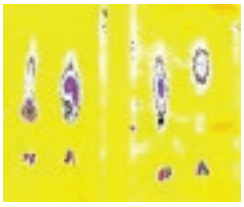
Electrophoresis of Alfalfa Extract *(**N** = normal, **A** = activated)*

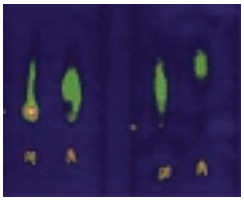


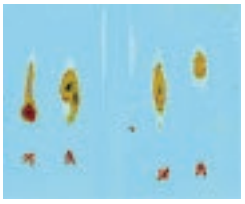
Extract A moves before and more than extract N (greater load/mass relation).

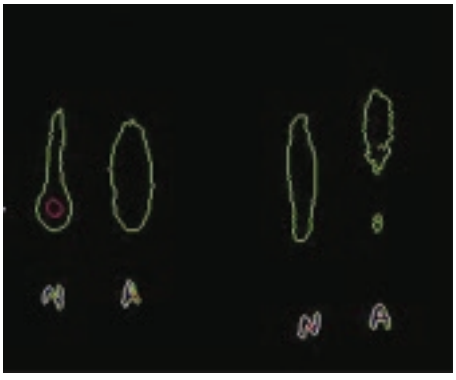


Left: substantial sample.
Right: less sample.
Tiny point in the left sample (edge): departure point (application of the sample).

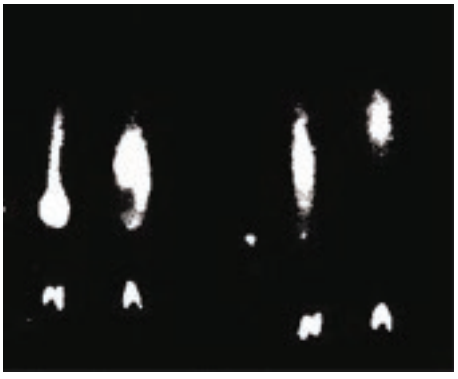








Representation in level curves.



Forced contrast to white and black.

The extract of activated alfalfa (A) has moved towards the positive pole, which indicates a greater negative load.

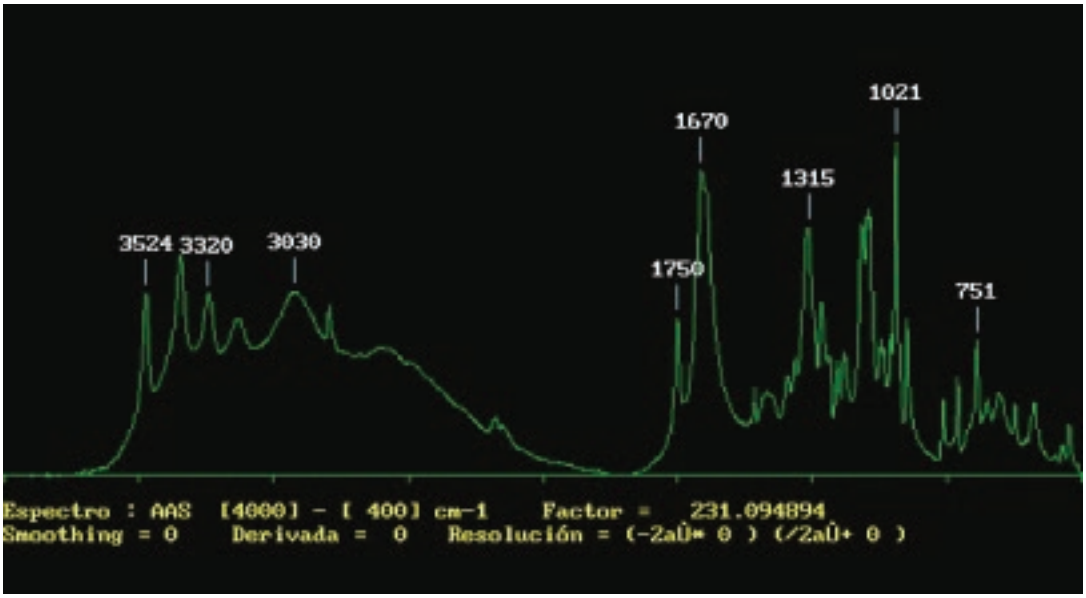
Extract A has a higher proportion of load/mass related molecules compared with the control extract (N).

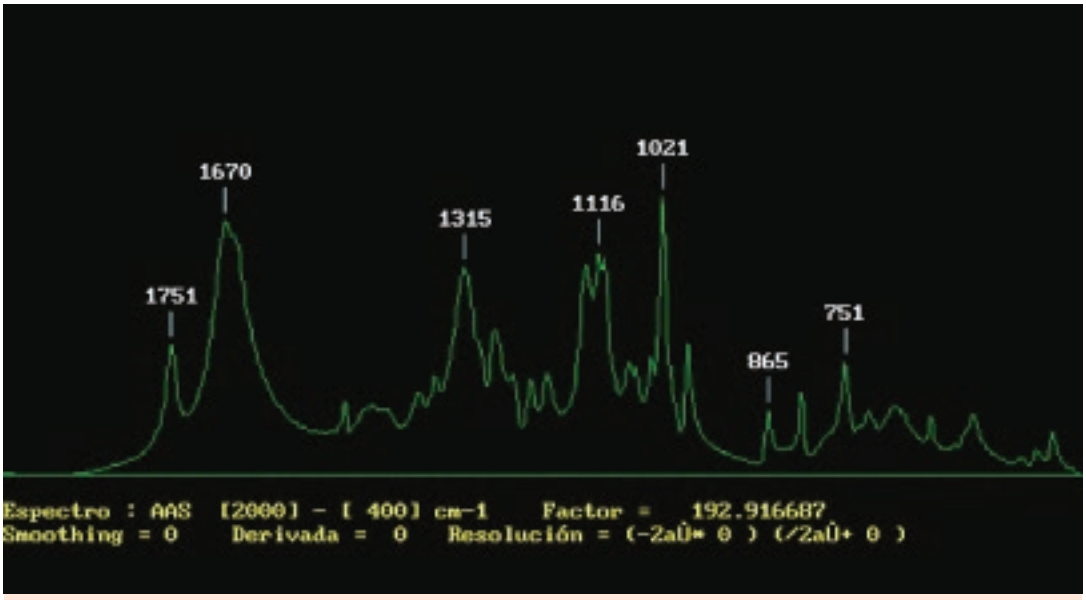
- ***Biophysical Analyses*** *- Infra-red*

The spectra of the infra-red analyses practised on molecules of activated and non-activated ascorbic acid and glycirrhizinic acid show that in both compounds the molecular activation has not broken any bond or formed any new bond.

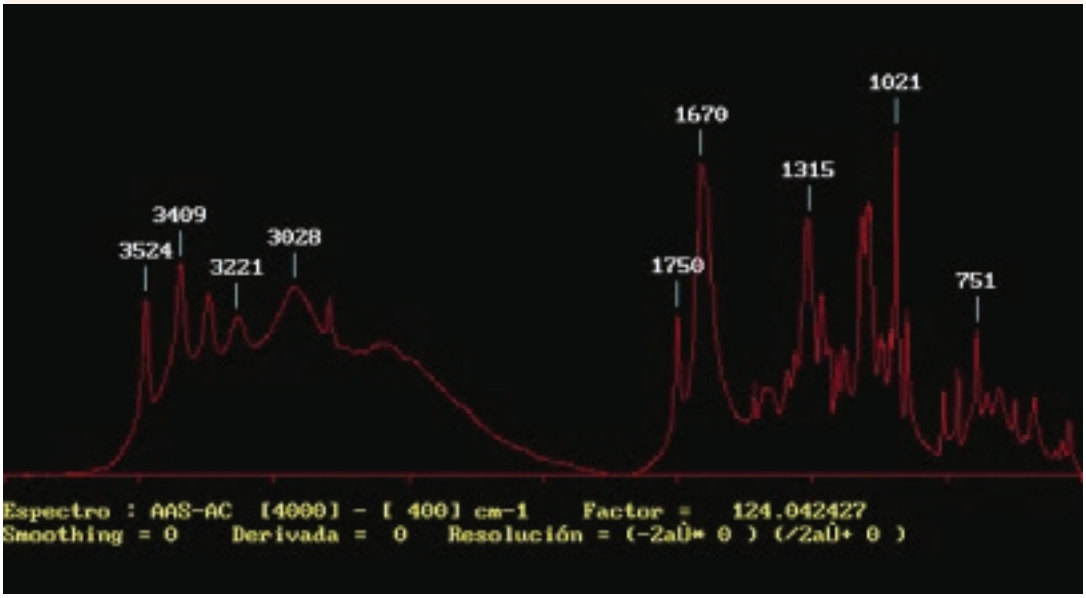
The study of these analyses confirms that no new compounds are formed.

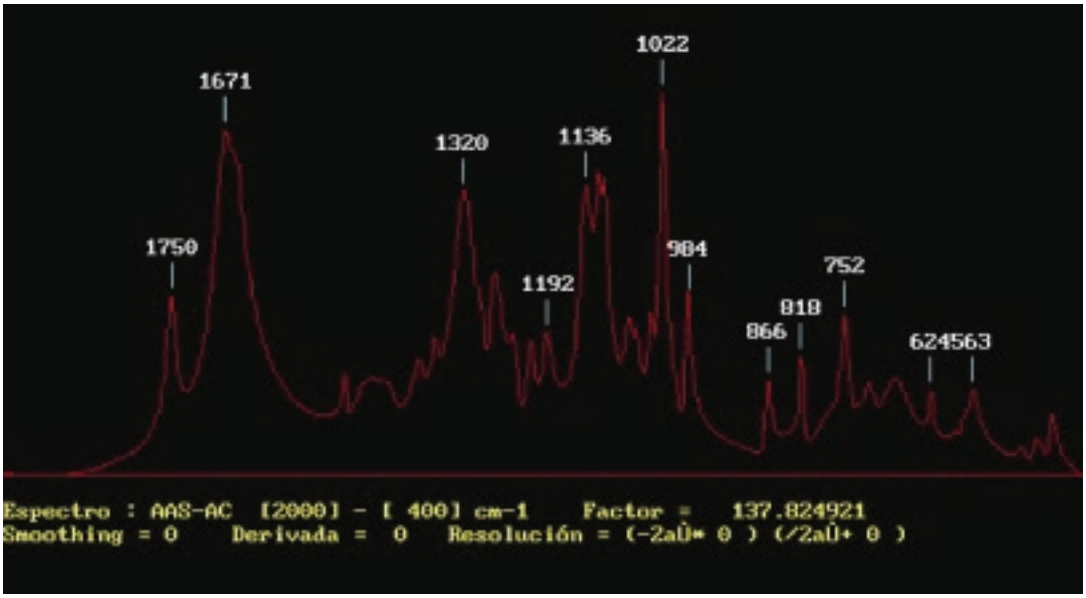
SPECTRA OF NON-ACTIVATED ASCORBIC ACID





SPECTRA OF ACTIVATED ASCORBIC ACID

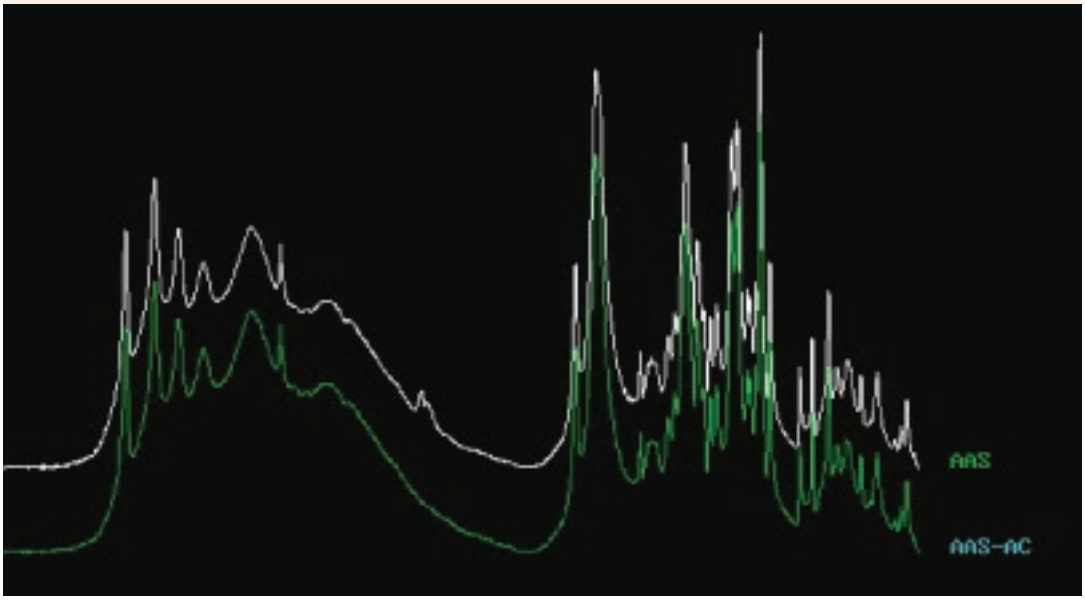




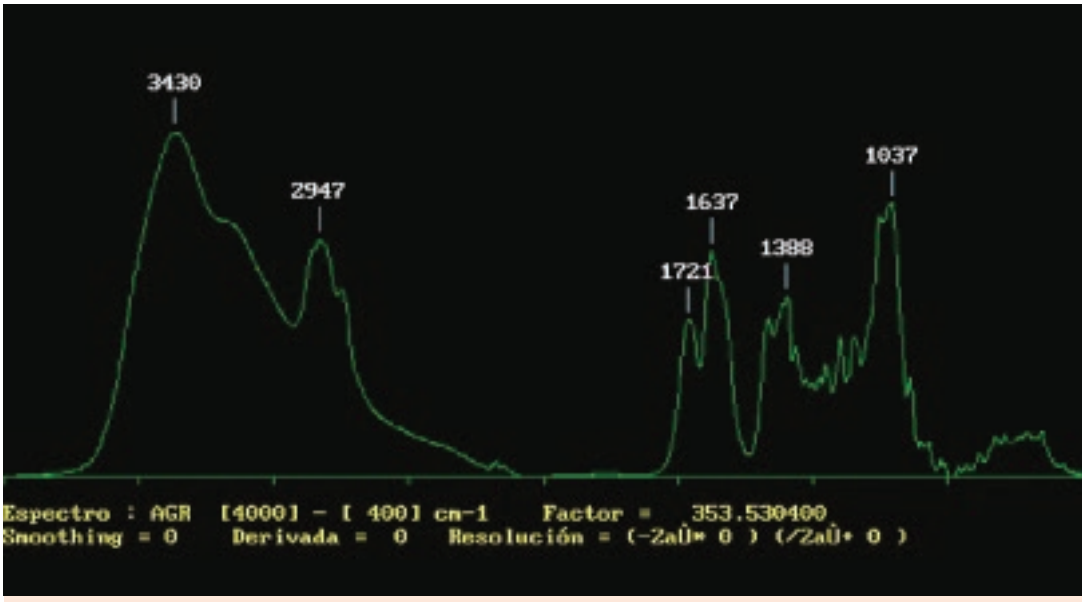
There are five higher peaks in activated ascorbic acid: 1670, 1315, 751, 624 and 563 cm-1.

These height changes represent respective percentage increases of: 40.9%, 44%, 40%, 40% and 50%. This increase in intensity from activation may possibly be related to the development of greater antioxidant capacity.

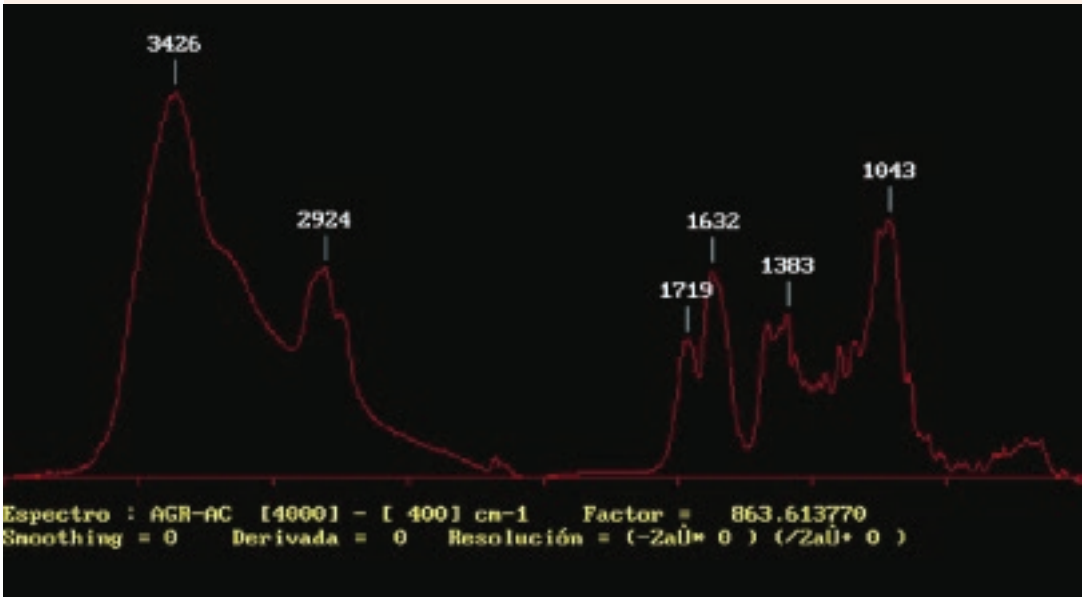
SPECTRA OF ACTIVATED AND NON-ACTIVATED ASCORBIC ACID (Superimposition)



SPECTRA OF NON-ACTIVATED GLYCIRRHIZINIC ACID

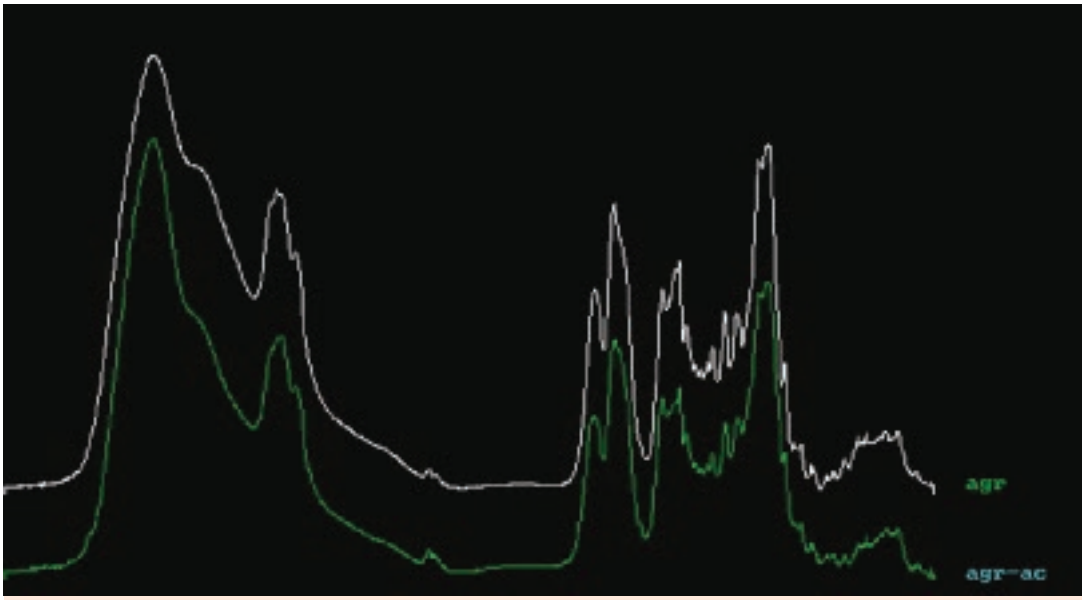


SPECTRA OF ACTIVATED GLYCIRRHIZINIC ACID



An increase of 13.33% is observed at peak 3430 in activated glycirrhizinic acid, in comparison to non-activated glycirrhizinic acid.

SPECTRA OF ACTIVATED AND NON-ACTIVATED GLYCIRRHIZINIC ACID (Superimposition)



Antiviral activity of activated Glucosamine against HSV Virus Type 2 in Rabbit Cornea

Herpes simplex virus is an important aetiological agent in eye infection and is the main cause of blindness in industrialised countries. Approximately 40% of all patients experienced one or more relapses before two years.

Herpes virus type 1 (HSV-1) causes most facial and lip infections in adults and herpes virus type 2 (HSV-2) causes most genital diseases.

Eye diseases caused by HSV-2 virus (conjunctivitis, corneal ulceration, iritis, etc.) are more severe and prolonged than eye diseases caused by HSV-1 virus.

Clinical Studies of Rabbit Cornea

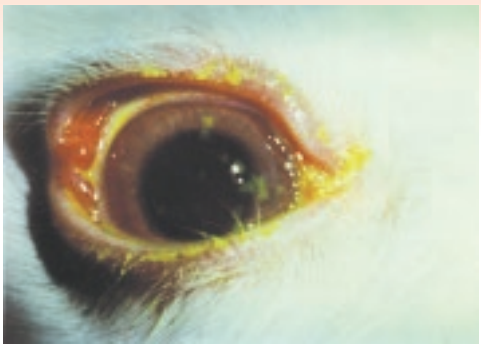
Activated glucosamine in a 10% aqueous solution was the compound used to study the effect of molecular activation on antiviral activity.

The compound control was Acyclovir (Zovirax) at a concentration of 3%, with the addition of DMSO to obtain an aqueous suspension (*See photographs on page 23*).

Average Results of Keratitis (in viral colonies)

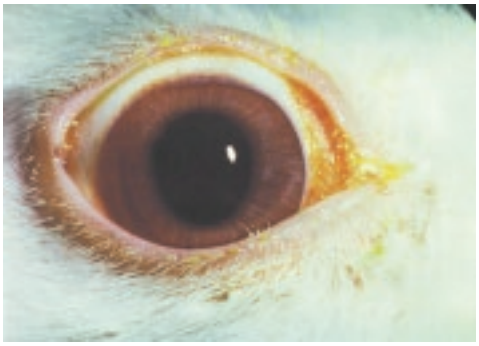
	4 days	*14 days*
Non-activated Glucosamine	***2***	***4***
ACTIVATED Glucosamine	***0,5***	***0***
Acyclovir	***0,5***	***0***
Control	***3***	***4***

Analysis of the trigeminal nerves revealed a complete absence of virus in the activated glucosamine therapy; this was not the case with Acyclovir, where viral colonies were detected, as this product is less penetrable. (Ophthalmological Act 67, 55-60 (1989)).



Control of HSV-2 virus
with non-activated glucosamine
4 days after viral infection.

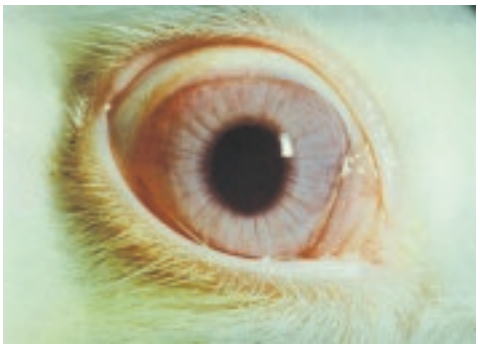
Control of HSV-2 virus
with non-activated glucosamine
14 days after viral infection.



Treatment of HSV-2 virus
with activated glucosamine
4 days after viral infection.

Treatment of HSV-2 virus
with activated glucosamine
14 days after viral infection.

Treatment of HSV-2 virus
with Acyclovir
4 days after viral infection.



Treatment of HSV-2 virus
with Acyclovir
14 days after viral infection.