



## Evaluation of the efficiency of the treatment applied to giant bamboo strips with pyroligneous acid against powder-post beetle

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### ABSTRACT

Despite numerous economic and environmental benefits of bamboo, its greater use is still hampered by the limited natural durability of most species. Among the products aiming to improve the protection of bamboo the pyroligneous acid occupies outstanding position. In this research work, bamboo splits were cooked in pyroligneous acid, at three different dilutions (10, 20 and 30%) at five time intervals (15, 30, 60, 120 and 180 minutes), in order to protect against deterioration by powder-post beetle. During 20 weeks, bamboo strips were visually inspected and having their mass loss measured, as well as readings of the ultrasonic pulse velocity (UPV) parallel to fibers. Paired t-test indicated that UPV decreased meanwhile strips mass increased; however it should be considered the simultaneous effect of the relative humidity changes in the period.

### KEYWORDS

*Dinoderus minutus*, borer, biodeterioration, alternative material, *Dendrocalamus giganteus*

### Introduction

*Dendrocalamus giganteus* Munro, one of the giant bamboo species, is suitable for constructions and furniture applications in the region southeastern of Brazil. Despite all the economic and environmental advantages, the use of bamboo is impaired by the low durability of the most of species. Fungi, bacteria and insects are responsible for bamboo deterioration, and powder-post beetle attack provokes a most serious economic damage. Thus, it becomes a challenge to search for appropriate bamboo protection employing natural or chemicals treatments and at the same time without provoking significant environmental impacts. Considering economic and environmental aspects pyroligneous acid can be a viable option for bamboo treatment. So, researches on efficient methods to protect bamboo against powder-post beetle attack are extremely important, aiming to increase the durability and acceptance of bamboo.

In Brazil, *Pinus* and *Eucalyptu* plantations were not enough to supply the demand for wood for construction, furniture, pulp and paper production, coal and energy for industrial purposes. In the other hand, bamboo shows characteristics suitable for wood replacement for several purposes, mainly due to its high growth rate. Recently, the Brazilian Government enacted the Law 12484 (2011) aiming to encourage native bamboo planting in family farms. Therefore, it is expected the forest sector, at a first step, will show interest in bamboo plantations.

Among the many possible uses of bamboo the application on glued laminated production stands out. The technology associated to glued laminated bamboo (GLB) is an appropriation of laminated wood, which for over a century has been developed and applied in the United States and Europe, designed for applications in construction, industry, sports facilities, among others.

GLB allows reducing the influence of peculiar bamboo geometry, permitting wood replacement in several applications. According to Pereira & Beraldo (2008), GLB is the most promising application of bamboo for several products as flooring, boards etc.

Unlike most of trees, bamboo does not produce toxic substances during its lifetime (Liese, 2003), and its degradation is caused by biological agents as fungi, bacteria and crustaceans (Highley, 1999). The order Coleoptera is the most important, including the families Anobiidae, Buprestidae, Scolytidae, Cerambycidae, Platypodidae, Lyctidae and Bostrychidae.

*Dinoderus genus* is responsible for causing serious economic damage to agricultural activities. Matoski (2005) reported the intense proliferation of *D. minutus* in stored wood veneers from tropical trees. After *D. minutus* attack bamboo and dry wood deteriorate, with significant economic impacts on the construction and furniture (Ahmed & Zulfiqar, 2006).

*D. minutus* exhibits cosmopolitan habits frequently developing in cut bamboo. This insect life cycle, distribution, food habits, growth and development are strongly related to prevailing environmental conditions (Garcia & Morrell, 2009). Under proper development conditions, populations of powder-post beetles cause considerable damage to the stored bamboo, since their life cycle occurs inside the material.

Powder-post beetle open galleries for oviposition and, soon after, entries were closed with the own dust generated during drilling (Matoski & Rocha, 2006). According to Garcia & Morrell (2009), eggs are elongated (180/220 microns x 810 microns) and are mainly deposited in metaxylem, which are the bamboo's vessels of larger diameter, located in the inner layers of the culm wall. The smaller diameter vessels observed in some bamboo species may partially explain its high natural resistance against powder-post beetle.

Adult specimens of *D. minutus* have crepuscular habits, being more active in low light conditions (Matoski & Rocha, 2006). The photoperiod is therefore a determining factor in the level of bamboo deterioration by powder-post beetle and this fact should be considered in projects of biological insect control and in the stored bamboo preservation.

Some aspects regarding the powder-post beetle creation are essential to facilitate the access to a large number of specimens in a short period of time:

Temperature - Garcia & Morrel (2009) pointed to the critical temperature of 34 °C for *D. minutus's* eggs developing; for higher temperature the hatch does not occur. Powder-post beetle develops from egg to adult in 53.8 days at a constant temperature of 30 °C. In fact, temperature strongly influences the development of insects and may increase or decrease its life cycle.

Photoperiod - is another extremely important factor in the powder-post beetle development, given the influence that this

factor has on its activity. The absence of photoperiod enhances the insect's activities, which may also accelerate its life cycle. Thus, deterioration tests can be faster when carried without photoperiod.

Natural cycle life – *D. minutus* management is enhanced by increasing environmental temperature, provoking population's increase and the natural behavior searching sex partners and new sources of food and place for shelter.

Insect's management – Bamboo compounds provides essential nutrients for powder-post beetle's development, being a substrate of easy access. It is suggested to employ more than one growth box.

Bamboo has great potential in construction, since the buildings are designed, built and properly maintained. However, some of these aspects are often neglected in construction, allowing the attack of insects and fungi (Nunes, Nobre and Saporiti, 2000).

Among the many ways to increase the wood and bamboo durability stand out an appropriate management, from cutting to final disposal, as well as the treatment with preservative solutions. The preservative treatment should be appropriate to the final bamboo application in construction, furniture or for other applications.

Different wood treatment procedures, also applied to bamboo, were high pressure and vacuum, dipping, diffusion and combinations of some of these techniques to apply the chemicals (Ibach, 1999).

Pyrolygineous acid, obtained by thermal decomposition of organic matter in the absence of oxygen, shows a great potential to protect the bamboo. Any organic matter such as wood and agricultural waste, during burning, three phases take place: a solid - charcoal, a gas (non-condensable gases) and which, by condensation becomes liquid - the pyrolygineous fraction, which also gets creosote, after settling.

Most of the time, the intense insect attack fully degrades bamboo, reducing its mechanical strength. Thus it would be interesting to survey the deterioration development over the time. Non-destructive testing (NDT) has been used as an alternative to classical destructive tests to assess the material's decay when exposed to weather conditions. This method is based on the propagation of high-frequency sound waves through the material as steel, concrete and wood. Ultrasonic pulse velocity (UPV) depends on the material's voids or discontinuities, which may indicate defects caused by deterioration. For example, NDT's application allows evaluating the characteristics of a material, as concrete, without changing their end-use capabilities (Lorenzi, Caetano, Drum and S. Filho; 2003; Oliveira, Candian, Luchette, Calil Jr, Salles; 2003).

### Methodology

This experimental part of this project was carried out in the Material Testing and Structures and Postharvest Laboratories, of the Faculty of Agricultural Engineering, at State University of Campinas (Unicamp).

Pyrolygineous acid and *D. giganteus* strips were provided by Oré Brazil Company, located at Campo Alegre - SC, through a partnership with Federal University of Santa Catarina (UFSC) and the Santa Catarina's Bamboo Association (BambuSC).

### D.minutus identification

Thirty individuals were collected and killed in a glass jar containing cotton soaked in chloroform, according to the methodology proposed by Gallo, Nakano, S. Neto, Pereira (2002) to identify bamboo powder-post beetle species. Three of them were fixed in entomological pin by double mounting procedure. Specimens were bonded with clear nail polish on the end of a small cardboard triangle and then introduced in the pin at the dorsal, ventral and lateral positions. A diaphragm

and a bamboo strip, both infested by powder-post beetle, were also studied to help understanding the insect's habits, which is an essential tool for the species determination. In addition, insects were collected for identification at Entomology Research Laboratory and Development Center of Plant Protection at São Paulo – SP.

### Growing specimens

Powder-post beetle specimens were taken from heavily infested diaphragms and of bamboo splits. Infested bamboos were kept without photoperiod placed in plastic boxes with nominal dimensions of 60.0 cm x 38.5 cm x 31.5 cm, with locking handles and without tight sealing. For each box it was adapted a mesh to prevent the free movement of specimens. Diaphragms and splits of bamboo provide amino acids, starch and simple carbohydrates, essential for the powder-post beetle development. Due to a high amount of parenchyma cells, bamboo's diaphragms and the inner wall of the culm have the most favorable conditions for *D. minutus* infestation (Liese, 1998). In the other hand, fiber bundles were very resistant to powder-post beetle attack. Therefore by discarding this region for the powder-post beetle growth, its stress decreases, then reducing the influence of this factor on the results.

### Number of insects required for tests

Nine hundred adults of *D. minutus* were removed from the breeding box and placed in contact with bamboo splits in the first week of the experiment. The German technical standard DIN EN 20-1 (1992) refers specifically to *Lyctus brunneus* (Stephen) (Lyctidae), which determines that the samples must be exposed to the action of eight individuals: four females and four males (Matoski, 2005). However, the sex determination of *D. minutus* is extremely complicated, as it must be done only in the larval stage. So, it was adopted a number of specimens that statistically guarantee the presence of four couples per specimen in deterioration tests. A binomial distribution, which returns the probability "p" of the existence of a number "x" of successes from "n" number of attempts, was employed. From a population of 898 powder-post beetle, the probability of finding 419 (or less) specimens of the same sex is 2.4%, which can be considered as statistically negligible. Specimens were then collected with a damp thin brush to avoid injuries by the use of a forceps. Less pigmented, morbid or accidentally injured insects were discarded. In the first six weeks, dead insects were replaced to ensure the presence of the originally proposed number of powder-post beetle for this period.

### Conditions for creation and management of borers

Tests were carried on plant growth camera with controlled atmosphere environment (Conviron, model EF7) and temperature, which are determining factor for the powder-post beetle development (Garcia & Morrel, 2009). Equipment was maintained at 26 °C with air circulation. Experiment was conducted in a room with temperature of (20 ± 2 °C) and controlled relative humidity (65 ± 5%) providing adequate ventilation. Powder-post beetle are more active in dark conditions being subjected to photoperiod only during the management phase.

### Specimen's mass determination

Mass powder-post beetle variation is a possible parameter for assessing the bamboo deterioration, since insect's life cycle occurs in practically all the bamboo. As recommended by Acda (2008), five groups with ten insects each of were separated. Masses were measured in analytical balance with a sensitivity of 0.0001 g. Average insect's mass was estimated as 3.0 ± 0.6 mg.

### Bamboo strips

Giant bamboo culms, acquired by the Oré Brazil Company from various farmers, were stored in the internal environment of the factory. Culms were cut by half in the longitudinal direction (parallel to the fibers). Innermost layer (rich in starch) and the outermost bamboo (bark, rich in bundle fibers) were removed. Then, strips were machined again to adjust to the measures as required by the deterioration test.

### Specimen's preparation

Bamboo splits were selected, according to the following criteria:

- Absence of faults cuts or wears which could influence the evaluation of the deterioration by powder-post beetle;

- Absence of deep stains, indicating preliminary bamboo colonization by fungi;

- Size: 20.0 cm x 2.0 (parallel to the bark) x 1.5 cm in the "radial" direction (transverse to the bark).

Ultrasonic pulse velocity (UPV) parallel to bamboo fiber is about 4500 m/s (similar to those of the wood). Resonance transducer's frequency was set at 45 kHz, so average wavelength was about 10 cm. The presence of knots in the specimens does not interfere in the UPV evaluation, however, it is known that this strip's region would more likely to suffer the powder-post beetle attack, due to the particular anatomical elements distribution, showing a higher starch content.

Due to the industrial process, bamboo strips were mixed from several parts of the culm. However, deterioration can be more intense at the specimens from the bottom region, due to their higher starch content. Considering the strip's thickness (1.50 cm) probably strips were taken from this region.

### Bamboo strip's treatment and preparation

Bamboo strips treatment was similar to those empirically used by Oré Brazil Company. Aqueous solution of pyroligneous acid at 30% concentration, were heated to 100 °C for 3 h.

### The variables used in the treatment were:

Soaking time (15, 30, 60, 120 and 180 min);

Dilution of the pyroligneous acid in water (0%, 10%, 20% and 30%).

Specimens were divided into groups of 30 splits and placed in a flask of 3 L. In another flask, pyroligneous acid solution was heated at 98 °C, and the heated solution was carefully poured in the first flask. At predetermined time intervals (15, 30, 60, 120 and 180 min), 5 splits were collected for each solution dilution (0%, 10%, 20% and 30%), totalizing 100 treated splits, besides 5 untreated ones (control). Due to the solution evaporation, the second flask was filled with pyroligneous acid solution to completely cover the strips. One of the treatments was conducted only with boiling water to check for the diluent effect in the treatment under consideration. Strips were then dried in a protected environment for 24 h.

Next, strips were marked on their transverse section, in the middle portions to the thickness and width, to facilitate the positioning of electroacoustic transducers for measuring the propagation time of the ultrasonic pulse, for UPV evaluation.

### Bio-assay - DIN EN 20-1

The evaluation of the bamboo deterioration by powder-post beetle was based on the European DIN EN 20-1 (1992) (Deuscht Verlag GmbH) norm. However, this standard is about the performance of wood preservatives against *L. brunneus* (Ste-phens) (Lyctidae) attack.

### Initial specimen's data

Splits were acclimated for one week in the Convicon equipment, prior starting the deterioration test. Specimen's mass was obtained in an analytical balance with a sensitivity of 0.0001 g. Specimen's bulk volume was calculated by the product of their average dimensions on tangential, radial and longitudinal directions.

### Deterioration box

After setting the Convicon equipment for a week, strips were placed in transparent glass box, with nominal dimensions of 16 cm x 25 cm x 35 cm. In the housing cover a mesh was adapted to prevent the exit/entrance of insects.

### Laboratory tests

To verify the effect of the different treatments mass and UPV across the strips were measured at the beginning of the bio-assays and after 4, 8, 12, 16 and 20 weeks of exposure to the powder-post beetle attack.

### Visual assessment

The condition of the specimens was visually classified from the criteria set out in Table 1.

**Table 1. Strip evaluation by visual observation.**

Categorie	Note	Criteria for classificacion
Intact	1	Absence of insect attack
Atacked	2	Marks of attack without insect penetration.
Penetrated	3	Presence of adult insect within the strips

### Ultrasonic pulse velocity (UPV)

UPV was obtained by calculating the ratio between specimen's length and the propagation time of the ultrasonic pulse in the longitudinal direction of bamboo strips (parallel to the fibers). A Steinkamp BP-7 device, featuring exponential section transducers of 45 kHz of resonant frequency, was employed. Transducers were positioned in the middle portion to the thickness and width of the strips (Espelho & Beraldo, 2008). The hypothesis was that the occurrence of defects in the strips, due to the powder-post beetle attack would increase the propagation time of the ultrasonic pulse, thus decreasing the UPV. The monitoring of the UPV's magnitude over the exposure time is an important tool to detect the strip's deterioration due to the insect activity, without having to destroy the infested material.

### Mass loss

Strip's mass was read in analytical balance (sensitivity of 0.0001 g) and sought to compare the variation between the initial and final specimen's mass after several periods of exposure to the powder-post beetle. The hypothesis was that the strip mass variation could be attributed to the larvae action, or due to the action of the second generation of *D. minutus*.

### *D. minutus's* mortality

During the first six weeks of the test dead powder-post beetle were collected and replaced by new specimens obtained directly from the boxes growth.

### Statistical analysis

Data were subjected to a series of analyzes using the Statistical Analysis System (SAS).

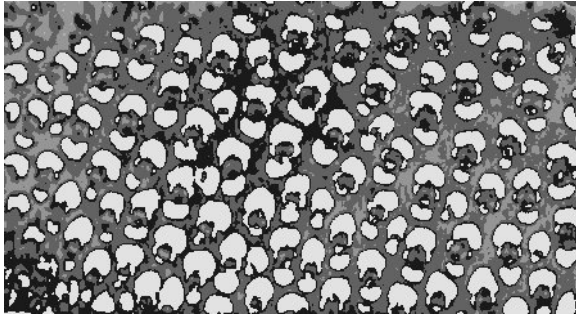
Analysis of variance (ANOVA) was applied to compare the contrast between two treatments and averages were compared by Tukey's test at the 95% statistical probability level. Analyses were carried by comparing the changes on strip's mass and on the UPV, according to the pyroligneous acid (PA) dilutions in water (0%, 10%, 20% and 30%) combined with soaking strips duration (15, 30, 60, 120 and 180 minutes)

### Results and Discussion

A major factor influencing the deterioration tests results refers to prior knowledge of the culm characteristics from which strips were produced. The bamboo specie, the culm age, the culm region of which specimens were obtained, the date of cutting (season of the year), and the way of drying the culms, exert great influence on intensity of the powder-post beetle attack. However, in this research work there was no information about these parameters, because bamboo was directly supplied by the industry.

Another important aspect is how strips were manufactured. As reported before, it was initially removed two strip's layers in order to obtain specimens with rectangular cross section. The first internal ones would be the most vulnerable to insect attack, for two reasons: higher starch content and large vessels which were the most favorable place for powder-post beetle's egg deposition (Garcia & Morrel, 2009). The second removed layer, rich-

er in bundle fibers and with vessels of smaller diameter, is one of the most resistant to powder-post beetle attack. Thus, when strips were manufactured it creates a type of bamboo mechanically improved, therefore, showing a better performance against powder-post beetle when compared to the natural bamboo. According to Liese (2003), the content of the bamboo fibers shows high variability ranging from 10% (inner layer) to 70% (outer layer). However, in this investigation average fiber content was about 30% (Da Silva & Beraldo, 2012), indicating, theoretically, a higher resistance to powder-post beetle attack (Figure 1).



**Figure 1 – *D. giganteus* cross section modified by image's software.**

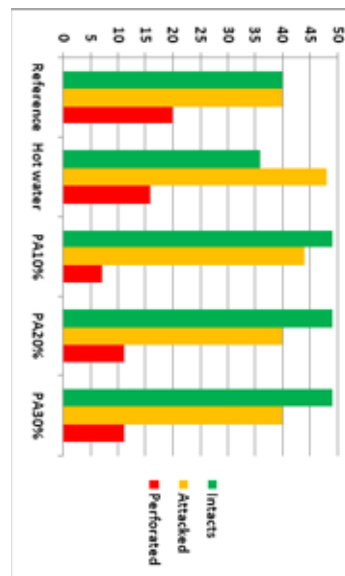
#### Visual assessment

The used light intensity induced the powder-post beetle to seek for shelter quickly. Considering that the opening galleries in bamboo are made by groups of two or more individuals, it is reasonable to assume that the insect preferably have infested the strips showing previous cavities made by other individuals.

Strips with signs of attack, but without evidence of powder-post beetle penetration, could indicate their perception of the presence of toxic compounds. However, there was no significant difference in the magnitude of the insect attack on reference strips when compared to those treated ones. Garcia & Morrell (2009) explained that individuals of *D. minutus* penetrate the bamboo preferably by cutting area or by cracks, or even by regions where the branches were removed at the knots region. Nevertheless, in this investigation, all of the bamboo surfaces were fully exposed, and it was observed that the preference of powder-post beetle was mainly at the node region. Probably, this fact is due to the higher starch content and the less mechanical resistance, as well as due to the lower fiber concentration in this region. Then, powder-post beetle penetration is facilitated by the crosslink among bamboo anatomical elements.

From 105 strips, 46.67% (49 strips) remained in perfect condition, denoting no signs of penetration or visible insect attack; 42.86% (45 strips) showed signs of attack and only 10.48% (11 strips) denoted powder-post beetle penetration (Figure 2).

The amount of intact strips did not show dependence of pyrolygneous acid (PA) solution concentration employed, but the amount of intact strips was higher when compared to the reference or those treated only in hot water solution. Concerning the perforated strips, it was observed that all of the PA concentrations employed reduced their numbers by a half when compared to reference ones.



**Figure 2. Bamboo splits percentage. PA (Pyrolygneous acid). Influence of relative humidity in the mass of specimens**

The objective of this analysis was to determine whether the variations of bamboo strips mass and the UPV across the specimens would be related to the relative humidity changes, which was a non-controlled parameter during the tests. Pearson correlation coefficient pointed to strong relationship between mass change and the relative humidity ( $p = 0.79$ ), which means that the split mass variation is hardly explained by the increase in relative humidity. However, the relationship between UPV and relative humidity was small and inversely correlated ( $p = -0.45$ ) indicating a tendency of UPV decreasing with increasing relative humidity. As for wood, UPV decreases with increasing moisture content up to the fiber saturation point (about 20% for bamboo), from which UPV remains constant.

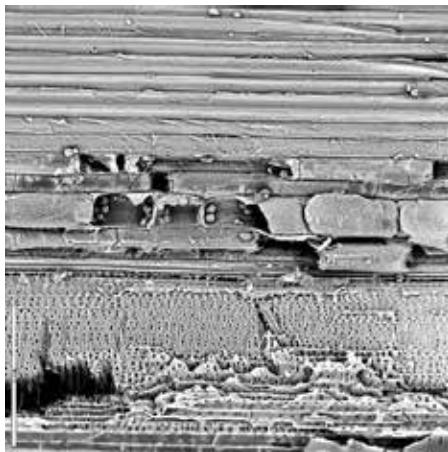
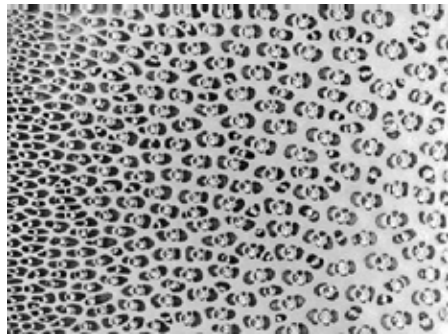
#### Ultrasonic pulse velocity - PV Effect of bamboo anatomy

Scatter plot of the correlation between bamboo fiber content and UPV pointed a reduction of 0% to 4%, for fiber content ranged from 20% to 55%. Analysis of scatter diagrams indicated that UPV changes were not correlated with bamboo fiber content.

UPV's results showed to be not consistent due to the transducer's characteristics ( $f = 45$  kHz), which restrained its application when the direction parallel to the bamboo fiber is considered, without substantial detection of bamboo strip deterioration (Figure 2a). Probably, more adequate results would be obtained by employing transducers of higher resonance frequency (500 kHz or 1 MHz). Then, it will be possible surveying bamboo strip deterioration by positioning the transducers at several points along the specimen's cross section. In this case, by considering UPV about 1500 m/s at the transverse section, wavelength will be of 1 mm ( $f = 1$  MHz) to 3 mm ( $f = 500$  kHz).

In reality, transducers emit a wave bundle which propagates faster in a heavier material, as bamboo fibers. Even if fiber's neighbor cellules of vessels and parenchyma could be deteriorated, the wave still propagates across the fibers, similarly the wave propagation in a steel bar, in a deteriorated reinforced concrete (Figure 3a). In the other hand, if transducers were applied to the bamboo's transversal direction several strip's regions could be evaluated, among which certainly occurs parenchyma cells (image's center) – the most probable deteriorated region (Figure 3b).





(a) Parallel (b) Transverse

**Figure 2. Bamboo anatomical directions. Fibers position.**

#### Effect of solution dilution

Tukey test revealed that there was significant differences among UPV on strips treated with PA at 10% to the others dilutions.

#### Effect of soaking time

Tukey test showed no significant differences among the different soaking duration with respect to the UPV changes.

#### Mass changes

Tukey test revealed that there was an only significant difference for PA20% when compared to the others treatments.

#### Effect of soaking times

Tukey test allowed the clustering of different soaking times in three statistically distinct groups. Treatments of 30 and 60 minutes of soaking were grouped as well as the treatments of 15 and 180 minutes. The third treatment group consisted of 120 minutes. Dunnett's test revealed that there was a significant difference between the mass strips changes of reference to those from 180 minutes of soaking time.

#### Conclusions

The proposed method for *D. minutus* growth was adequate for the purpose of this research. However, many adjustments must be made in testing, especially regarding to the relative humidity control, a crucial factor to be able to assess the strip mass variation of bamboo only due to the powder-post beetle attack.

Despite strips mass showed a slight increase (0.99%) in the period, it must be considered that this result was disturbed with the simultaneous strip mass loss due to the powder-post beetle attack, because strip mass increases after water absorption.

A large amount of intact strips (46.67%) compared to those with powder-post beetle penetration signs (10.48%) indicates that an effective bamboo strip protection regardless to the treatment. Thus, the 20 weeks period in which the deterioration test was conducted not allow to select optimal parameters for improving the bamboo strip treatment pyrolytic acid.

After 20 weeks, it was found that there was only a slight decrease (1.70%) in UPV. In addition, statistical analysis did not show differences in UPV on strips treated with different soaking times

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