



SLOVENSKI STANDARD
SIST EN 48:1996

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Wood preservatives - Determination of the eradicator action against larvae of Anobium punctatum (De Geer) (Laboratory method)

Holzschutzmittel - Bestimmung der bekämpfenden Wirkung gegenüber Larven von Anobium punctatum (De Geer) (Laboratoriumsverfahren)

Produits de préservation des bois - Détermination de l'efficacité curative contre les larves d'Anobium punctatum (De Geer) (Méthode de laboratoire)

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Ta slovenski standard je istoveten z: EN 48:1988

ICS:

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EUROPEAN STANDARD
NORME EUROPÉENNE
EUROPAISCHE NORM

EN 48

November 1988

UDC 674.048.4:620.193.87

Supersedes EN 48,
August 1976

Key words : Wood, Wood preservatives, Pesticides, Insecticides, Protection against the pest, Laboratory tests, Determination, Effectiveness limit, Anobiidae, Curative treatment.

English version

Wood preservatives;
Determination of eradicant action against
larvae of *Anobium punctatum* (De Geer)
(Laboratory method)

Produits de préservation des bois;
Détermination de l'efficacité curative
contre les larves d'*Anobium punctatum*
(De Geer) (Méthode de laboratoire)

Holzschutzmittel; Bestimmung
der bekämpfenden Wirkung gegen-
über Larven von *Anobium punctatum*
(De Geer) (Laboratoriumsverfahren)

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CEN

European Committee for Standardization
Comité Européen de Normalisation
Europäisches Komitee für Normung

Central Secretariat : Rue Bréderode 2, B-1000 Brussels

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Ref.No. EN 48:1988 E

B R I E F H I S T O R Y

This European Standard was drawn up by the Technical Committee CEN/TC 38 "Methods of test for wood preservatives", the Secretariat of which is held by AFNOR.

According to the Common CEN/CENELEC Rules, following countries are bound to implement this European Standard :

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0 Introduction

This European Standard specifies a laboratory method of test which gives a basis for the assessment of the eradicator action of a wood preservative against *Anobium punctatum*. It allows the determination of the lethal effect of a surface application of the preservative on a population of larvae already established in the test specimens.

The method simulates conditions in practice where a length of wood such as an affected stair tread is treated, which is still free from exit holes and in which certain of the faces are inaccessible, thus constituting severe test conditions.

This laboratory method provides one criterion by which the value of a preservative can be assessed. In making this assessment, the methods by which the preservative may be applied should be taken into account. It is further recommended that results from this test should be supplemented by those from other appropriate tests and, above all, by practical experience.

When using products which are very active at very low concentration, it is of great importance that suitable precautions be taken to isolate and separate as far as possible operations involving chemical products, other products, treated wood, all clothing and laboratory apparatus. Suitable precautions shall include the use of separate rooms, areas within the rooms, extraction facilities, conditioning chambers and special personnel training.

1 Object

This European Standard specifies a method for the determination of the eradicator action of a wood preservative against larvae of *Anobium punctatum* (De Geer).

2 Field of application

This method is applicable to:

- organic water-insoluble formulations, as supplied or as prepared in the laboratory by dilution of concentrates; or
- organic water dispersible formulations as supplied or as prepared in the laboratory by dilution of concentrates; or
- water-soluble materials, for example salts.

3 Principle

Insertion of larvae of *Anobium punctatum* into one or more sets of test specimens of a susceptible wood species.

After establishment of the larvae, treatment of these specimens by brushing or spreading of the preservative.

After the time necessary for the preservative to take effect, estimation of the mortality of the larvae compared with that in untreated control specimens.

4 Test material

4.1 Biological material

Anobium punctatum (De Geer) larvae.

4.1.1 Source of larvae. Obtain the larvae from cultures reared as described in annex D.

Cut up this wood and extract the larvae in an area separate from the test environments (4.3.1 to 4.3.3) so as to avoid the risk of introducing mites.

Prepare the storage blocks from Scots pine sapwood of dimensions 50 mm x 25 mm x 15 mm, each with 10 evenly spaced holes (see 7.1) drilled into one of the wide longitudinal faces with the drill (4.3.4).

Before inserting the larvae into the storage blocks, keep them overnight in small glass receptacles.

Then sort the larvae into small, medium and large sizes. Do not use the large larvae, with a mass greater than 5 mg, for this test¹⁾.

The larvae shall be distributed as evenly as possible according to their mass. For example, for a single test the 216 larvae shall be distributed in 18 groups each of 12 larvae; the mean mass of the larvae in each group shall be approximately 3.5 mg¹⁾.

Examine all the other larvae under a binocular microscope and destroy those which are damaged or infested with mites, keeping only those that are in perfect condition.

Insert the small and medium larvae into separate sets of storage blocks, placing each larva head first into a drilled hole.

Keep these filled storage blocks, holes uppermost, in glass containers covered with filter paper fixed with adhesive.

Keep the larvae in the storage blocks in the culturing chamber (4.3.1) for not less than 2 months before using them in a test.

4.1.2 Provision of larvae. Carefully split the storage blocks, extract the larvae and examine them under a binocular microscope. Destroy any larvae that show injury or mite infestation, or that do not respond by movement when touched. It is particularly important to avoid including mite-infested larvae (see annex C).

Keep those larvae that are between 2 mg and 5 mg²⁾ in mass and in perfect condition overnight, separate from one another, in clean receptacles in the culturing chamber (4.3.1). Then re-examine them, and again reject any which do not show normal movements.

4.1.3 Choice of larvae. The larvae used in the test shall be between 2 mg and 5 mg in mass, and the 12 larvae placed in each test specimen shall have a mean mass of approximately 3.5 mg¹⁾.

The numbers of larvae required are indicated in table 1.

4.2 Products and reagents

4.2.1 Pure paraffin wax, setting point 52 °C to 53 °C, for sealing the transverse and narrow longitudinal surfaces of specimens to be treated with solutions in which water is the continuous phase.

¹⁾ The mass may be judged by eye by comparison with larvae of known mass.

²⁾ Experienced operators can judge the sizes well enough by eye.

Table 1. Number of larvae and test specimens			
Type of test specimen	Number of preservative concentrations or methods of treatment	Number of test specimens	Number of larvae
<i>Treated specimen</i>			
Softwood	1	6	72
	2	12	144
	3	18	216
Hardwood	1	6	72
	2	12	144
	3	18	216
<i>Untreated control specimen</i>			
Softwood	1	3	36
	2	3	36
	3	6	72
Hardwood	1	3	36
	2	3	36
	3	6	72
Total for one preservative with one concentration and method of treatment			216
Total for two preservatives with one concentration and method of treatment			360
Total for three preservatives with one concentration and method of treatment			576

4.2.2 *Pure gelatin*, for sealing the transverse and narrow longitudinal surfaces of specimens to be treated with solutions in which an organic solvent is the continuous phase.

4.2.3 *Water*, distilled or deionized.

4.3 Apparatus

4.3.1 *Culturing chamber*, with air circulation and controlled at between 20 °C and 22 °C with a tolerance of ± 1 °C and at between 80 % r.h. and 85 % r.h. with a tolerance of ± 5 % r.h.

4.3.2 *Laboratory work area*, well ventilated, where treatment of the test specimens is carried out³⁾.

4.3.3 *Testing chamber*, ventilated and air conditioned, and controlled at between 20 °C and 22 °C with a tolerance of ± 1 °C and at between 70 % r.h. and 75 % r.h. with a tolerance of ± 5 % r.h.

4.3.4 *Drill*, provided with a bit capable of drilling cylindrical or conical holes as specified in 7.1.

4.3.5 *Protective gloves*.

4.3.6 *Ordinary laboratory equipment*, including two analytical balances.

4.3.7 *X-ray apparatus* (if desired) with tungsten target and beryllium window, with voltage and current continuously variable in the ranges:

voltage: 10 kV to 50 kV

current: 0 mA to 15 mA

5 Sample of the preservative

The sample shall be representative of the product under test.

6 Test specimens

6.1 Species of wood

The reference species are:

Scots pine (*Pinus sylvestris* Linnaeus)

Beech (*Fagus sylvatica* Linnaeus)

Additional tests may be made using other species but if so, this shall be stated in the test report.

6.2 Quality of wood

Use only sound wood, straight-grained, without knots. For Scots pine only, sapwood with a low resin content shall be used and for beech, wood free from 'red-heart'.

Cut the test specimens from wood of average growth rate (2.5 to 8 annual rings per centimetre for pine, 2 to 6 annual rings per centimetre for beech).

The proportion of summer wood in the annual rings shall not exceed 30 % of the whole in the case of pine.

The wood shall neither have been floated nor subjected to any chemical or heat treatment⁴⁾. It shall be air dried and shall not have been stored for more than 5 years.

6.3 Provision of test specimens

Cut the test specimens from scantlings or beams so that, on the transverse cross section, the annual growth rings form an angle of $45 \pm 10^\circ$ with the longitudinal faces (see figure 1).

The test specimens shall be planed very carefully.

Avoid using test specimens from the butt or crown of the tree. Take the specimens for a test from three trees. Test specimens taken from the same tree shall be similar; they are considered such when the regions from which they are taken in the direction of the grain of the wood are not more than 1 m apart.

See figure 2 for the selection and distribution of test specimens.

6.4 Dimensions of test specimens

The nominal dimensions of each test specimen, measured at 12 % (*m/m*) moisture content, shall be 100 mm x 50 mm x 30 mm.

The total surface area of the faces exposed to treatment is theoretically 100 cm².

Check the size of each specimen to determine the actual area treated. Allow for any possible encroachment of the sealing compound on to the treated faces of the specimen.

³⁾ Proper safety procedures for handling flammable and toxic substances shall be followed.

⁴⁾ Gentle artificial drying at below 60 °C is, however, permitted.

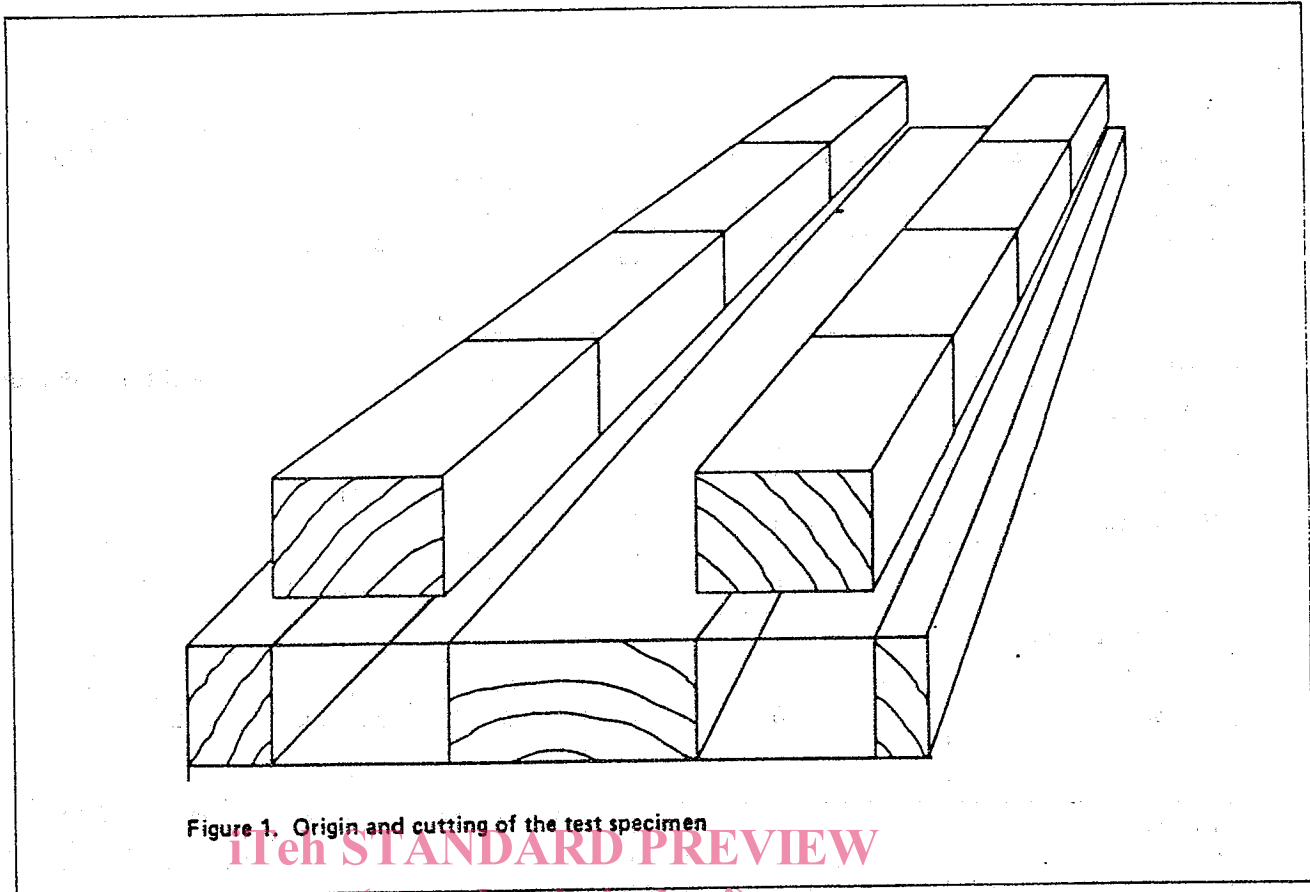


Figure 1. Origin and cutting of the test specimen

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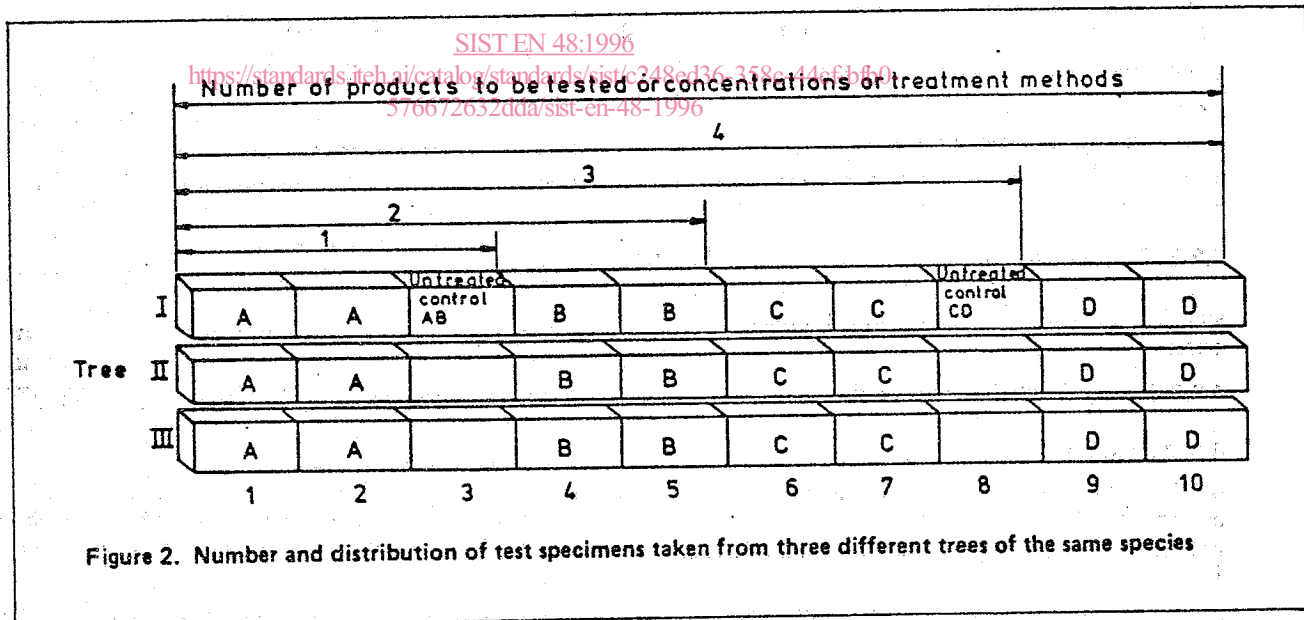


Figure 2. Number and distribution of test specimens taken from three different trees of the same species

6.5 Number of test specimens

Use for each preservative, each concentration, each method of treatment and each species of wood:

- six treated test specimens (two per tree);
- three untreated control specimens (one per tree).

If the examination involves, at the same time, several preservatives, concentrations or methods of treatment, three untreated control specimens are sufficient for each series of 12 treated test specimens.

7 Procedure

7.1 Exposure of test specimens to insects

Keep all the test specimens in the culturing chamber (4.3.1) for 2 weeks before drilling the holes to take the larvae.

Drill⁵⁾ (4.3.4) six cylindrical holes approximately 5 mm deep or six conical holes approximately 7.5 mm deep in the two transverse cross sections of each test specimen.

Drill a pattern of holes in two lines of three, 10 mm from the large faces of the test specimen with a distance between

⁵⁾ Ensure that the holes have smooth walls to prevent damage to the larvae.

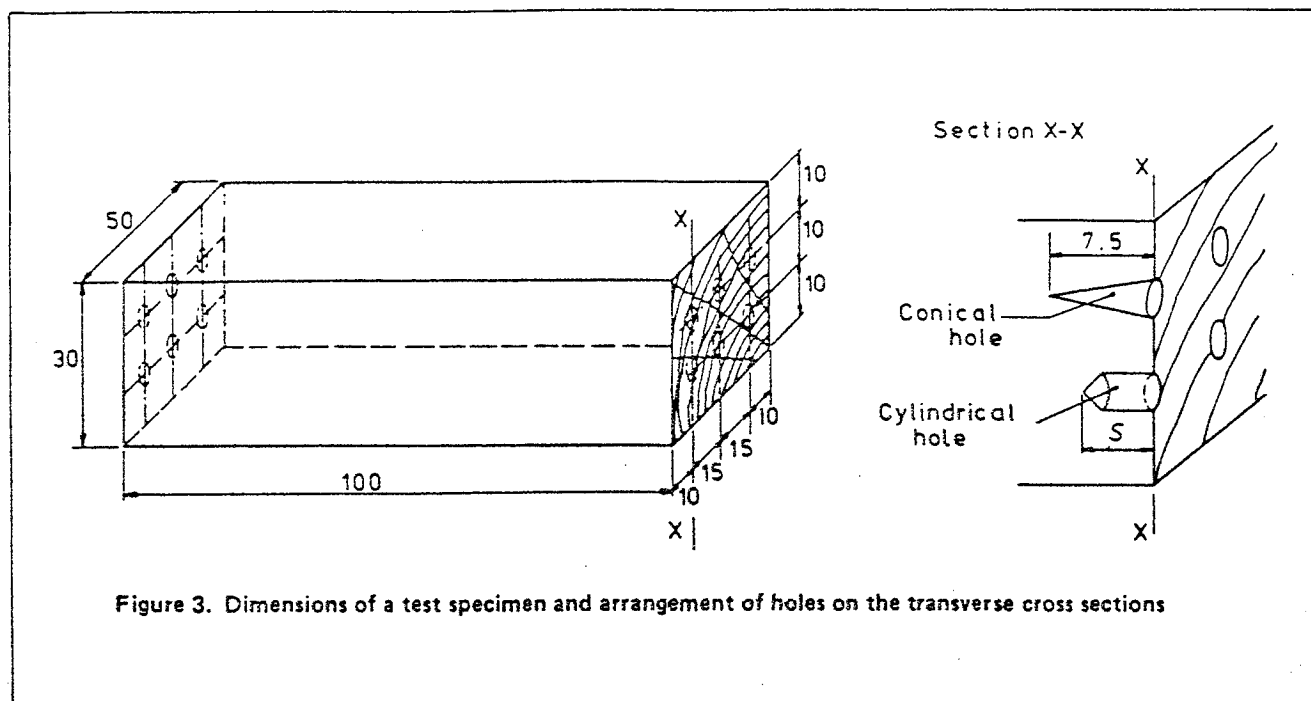


Figure 3. Dimensions of a test specimen and arrangement of holes on the transverse cross sections

the holes in the same row of 15 mm and a distance between the two rows of 10 mm (see figure 3).

Determine the diameter of the holes according to the mass of the larvae from table 2.

Insert the selected larvae head first into the 12 holes of each of the six treated test specimens and the three control specimens.

Mass of larvae	Approximate hole diameter
mg	mm
2 to 2.9	1.2
3 to 3.9	1.4
4 to 5.0	1.6

Seal the entrances to the holes by means of a glass plate and fix this to the test specimen by means of a narrow adhesive tape⁶⁾.

Keep the test specimens in the culturing chamber (4.3.1) for 12 weeks, placing them on their wide faces. During this period, the larvae should start tunnelling during the first 2 or 3 weeks. Replace any larvae which do not start boring.

Avoid any lowering of temperature for the stored larvae and those introduced into the treated test specimens⁷⁾.

If radiographic examination (4.3.7) is used, ascertain the position of the larvae so that their further tunnelling can be checked in later examinations.

7.2 Preparation of test specimens

7.2.1 Sealing of the non-treated surfaces. Remove the glass plate from the transverse cross section and seal the narrow longitudinal faces and the transverse cross sections as follows.

7.2.1.1 For tests with solutions in which water is the continuous phase. Use three coats of the pure paraffin wax (4.2.1) at about 100 °C, so that the first coat adheres closely to the wood and successive layers bond closely to one another.

7.2.1.2 For tests with solutions in which an organic solvent is the continuous phase. For tests with organic solvent-based preservatives, which would dissolve paraffin wax, use the pure gelatin (4.2.2). Apply the first coat with a 200 g/L aqueous solution at 40 °C; then, after a short period of drying, apply two further coats of 300 g/L aqueous solution at 50 °C.

7.2.2 Treatment of the test specimens

7.2.2.1 Preparation of treatment solutions

7.2.2.1.1 Solid water-soluble preservatives. Dissolve the preservative in the distilled or deionized water (4.2.3) to the predetermined concentration.

7.2.2.1.2 Liquid preservatives. If appropriate, use the preservative without further preparation other than any necessary stirring.

If the preservative is a concentrate, dilute the preservative with the diluent to the required working concentration, as specified by the manufacturer.

7.2.2.2 Treatment. Carry out the treatment in the laboratory work area (4.3.2). Apply the preservative to the

⁶⁾ It is also possible to insert the larvae in two stages. First, larvae are inserted in one end only of each test specimen and the specimens are left for 1 week with the ends containing the larvae uppermost. After 1 week, the test specimens are inverted and the larvae are inserted into the second end.

⁷⁾ Any drop in temperature encourages pupation, which interferes with the test. For biological reasons, the months from July to January are the most favourable for carrying out the tests.