PARTICLEBOARDS PREPARED WITH ADDITION OF COPPER SULPHATE – PART 1: BIOLOGICAL RESISTANCE

Ladislav Reinprecht – Zuzana Vidholdová – Ján Iždinský

ABSTRACT

Protection of particleboards (PBs) with inorganic water soluble biocides (e.g. copper salts) can be recommended for indoor occasionally moistened expositions – i.e. at a random effect of condensate water or at a water leaking from damaged pipeline – lasting a short time without damage of the adhesion connections between wooden particles and glue. This work investigates biological resistance of the one layer PBs prepared by addition of copper sulphate pentahydrate (CuSO$_4$.5H$_2$O) into melamine-urea formaldehyde (MUF) glue in the amounts of 0, 2, 6, 12 or 24 w/w %. MUF glues containing copper biocide had, in presence of hardener NH$_4$NO$_3$, a shortened curing time from 198 to 117 s. This biocide evidently supressed occurrence of bacteria on the top surfaces of PBs, the gram-positive bacteria Staphylococcus aureus by up to 76.5 % and the gram-negative bacteria Escherichia coli by up to 50.0 % – i.e. the activity of bacteria at using a 0.5 McFarland standard inoculum (1.5 $\times 10^8$ CFU/ml) drop in 48-hour tests from 0.17 to 0.04 $\times 10^8$ CFU/ml for S. aureus and from 0.18 to 0.09 $\times 10^8$ CFU/ml for E. coli. The copper biocide also increased up to 44.5 % the decay-resistance of PBs against the brown-rot fungus Coniophora puteana – i.e. the mass losses of the copper-modified PBs reduced at 6-week tests from 17.38 % to 9.64 %. However, the anti-mold efficacy of CuSO$_4$.5H$_2$O was a partly milder at reducing growth of the microscopic fungi Penicillium brevicompactum and Aspergillus niger on the top surfaces of PBs – i.e. the activity of molds was suppressed by up to 25.1 % and 31.5 %, when the molding degrees valued by the Standard STN 49 0604 reduced at 28-day tests from 2.67 to 2.0 for P. brevicompactum and from 3.17 to 2.17 for A. niger.

Key words: particleboards, copper sulphate, bacteria, molds, brown-rot fungi.

INTRODUCTION

Particleboards (PBs) are well known wood based composites (prepared from ~ 90 % of wood particles, ~ 9 % of glues, ~ 1 % of paraffin and other additives) applied typically in indoor exposures. Resistance of PBs to biodeterioration processes is affected by the species of wood(s), the type and amount of adhesive used as binder, the size and shape of wood particles, and composition and physical properties of the final board (IMAMURA 2013).

Generally, an increased biological resistance of PBs against bacteria, molds or decaying fungi is important only in such situations if they in interior expositions (the use classes 1 and 2 by EN 335 2013) must also withstand short time impact of additional wetting with achieving a moisture content over 20 % (e.g. at presence a condensate water during 1/2–14 days). PBs for dry indoor conditions must not be water-resistant and biologically-
resistant to bacteria and fungi (unlike of water-resistant plywood suitable for the use classes 3 and 4, etc.), however, in specific situations should be increased their biological durability applying convenient biocides (Kirkpatrick and Barnes 2006).

Increased resistance of PBs to bacteria, molds and decaying fungi is usually realised by their: (1) post-manufacture surface treatment (PMST), for example: - lamination with traditional impregnating papers (Vidholdová et al. 2015), - lamination with impregnating papers containing biocides (Hanraman et al. 2006), - painting with acrylic, epoxy and other coatings without or with biocides, - gassing with fungicides, e.g. with trimethylborate (Murray 1994) or supercritical carbon-dioxide fluid of organic fungicide “SCF-CO2” (Morrell et al. 2005); (2) in-process treatment (IPT) applying biocides during the production process: for example: - impregnation of wood particles with thermostable inorganic, heterocyclic or other fungicides (Reinprecht and Štefka 1989), - using special bio-active glues, - addition of fungicides into glues (Reinprecht and Perlac 1995), - addition of fungicides when applying a glue, when layering wood particles, or during pressing of particleboards.

The aim of this study was to search bacterial and fungicidal efficacy of the well-known inorganic biocide “copper sulphate pentahydrate (CuSO_{4}.5H_{2}O)” used at preparation of one-layer PBs by IPT technology, in this experiment by adding it into the melamine-urea formaldehyde (MUF) glue.

Inorganic copper fungicides are commonly applied as copper oxide CuO, hydroxide Cu(OH)$_2$, carbonate CuCO$_3$, hydroxide-carbonate Cu(OH)$_2$.CuCO$_3$, or pentahydrate of copper sulphate CuSO$_4$.5H$_2$O. Wood and wooden composite treated with copper compounds have a green colour, so this can be their particular disadvantage from aesthetic views. Copper denatures enzymes of bacteria and fungi. It is active in particular against soft-rot fungi (Ray et al. 2010, Karunasekera et al. 2017). Green and Clausen (2003) at application of ammonia copper citrate into a solid wood found that the brown-rot fungi Coniophora puteana MAD 5 15 or Serpula lacrymans Bam Ebers 3 15 are copper-sensitive. On the other hand, more species of tested brown-rot fungi (e.g. Antrodia vailantii, Postia placenta, Wolfiporia cocus), which use the Fenton reaction at depolymerisation of cellulose, are tolerant towards copper-based wood preservatives as a consequence of creation the non-bio-active copper oxalate crystals, as well as other gene predictions. It is in a conformity with works of Hastrup et al. (2005), Schilling and Inda (2010) or Jenkins et al. (2014). Copper compounds are mainly used in water, amine or organic solutions, but in recent years also in the form of nano-copper – micronized copper particles (MCQ), which dimension is usually from 10 to 700 nm (Matsunaga et al. 2007, McIntyre 2010). Today is known that in presence of Cu$^{2+}$ ongoing oxidation process in polysaccharides and also in guaiacly-lignin to quinone-methides, which led to complex formation of copper with all wood components.

MATERIALS AND METHODS

Treatment of MUF glue with CuSO$_4$.5H$_2$O
The basic melamine-urea formaldehyde (MUF) glue was prepared in a molar ratio of formaldehyde and urea 1 : 1.15 with addition of 20 weight % of melamine. It was used as a 67 % water solution. The hardener of the MUF glue was ammonium nitrate (NH$_4$NO$_3$), and it was added as a 57 % water solution in amount of 2.5 w/w % (weight of solid hardener per solid weight of catalysed glue).

The copper-modified MUF glues contained copper sulphate pentahydrate (CuSO$_4$.5H$_2$O) in the amounts of 0 %, 2 %, 6 %, 12 % or 24 w/w % (weight of solid copper biocide per solid weight of catalysed MUF glue). MUF glues treated with CuSO$_4$.5H$_2$O had
a shortened curing time (Table 1), while their viscosity determined by Ford φ 4 mm was constantly 62 ± 1 s.

**Tab. 1 Curing time of the copper-modified MUF glues at 100 °C.**

<table>
<thead>
<tr>
<th>MUF glue</th>
<th>CuSO₄·5H₂O (weight /per solid weight of catalysed MUF glue)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0 %</td>
</tr>
<tr>
<td>Curing time [s]</td>
<td>198</td>
</tr>
</tbody>
</table>

**Particleboard’s preparation**

The 1-layer particleboards (PBs) with the area dimensions of 360 mm × 360 mm, the nominal thickness of 16.0 mm, and the density of 640 ± 15 kg.m⁻³ were prepared in a laboratory. For their production were used the following substances: (1) 10 parts – dry mass of wood particles (9.5 part from spruce and other softwoods, and 0.5 part from beech, oak and other hardwoods) used at 3.5 % EMC; (2) 1 part – dry mass of MUF glue (0.965 part of MUF glue, 0.025 part of hardener ammonium nitrate, 0.010 part of paraffin) used as 67 % water solution; (3) 0 to 0.24 part – dry mass of copper sulphate pentahydrate.

Wood particles were firstly homogenized in a rotary mixing device with copper-modified MUF glues, then loaded into a pre-pressing form, and finally pressed. Pressing process was performed in a laboratory press CBJ 100-11 according the three stage pressing diagram at a temperature of 210 °C, a maximal specific pressure of 5.33 MPa, and a pressing factor of 14 s (IŽDINSKÝ et al. 2013). Totally one control type and four modified types of 1-layer PBs were produced (each variant in 3 pieces, so that altogether 15 PBs).

**Biological properties of particleboards**

From the control and modified PBs were cut samples for testing their biological resistance against bacteria, molds, and the brown-rot fungus *C. puteana*. Samples used for biological tests had a constant testing thickness of 7.0 mm which was achieved by cutting of pressed PBs at their half-height. Before individual biological tests the samples were conditioned for two weeks at temperature of 20 ± 2 °C and relative humidity of 65 ± 2 %, for attaining 12 ± 1 % EMC. Then the side and bottom surfaces of samples were 2 times pained with epoxy resin (CHS-Epoxy 1200, hardener P11) and their all surfaces sterilized 1 h with UV light.

**Bacterial test**

Anti-bacterial resistance of the top surfaces (50 mm x 50 mm) of PB’s was searched with the gram-positive bacteria *Staphylococcus aureus* ATCC-25923 and the gram-negative bacteria *Escherichia coli* ATCC-25922. Samples were cleaned with alcohol solution (8.8 : 1.2 mixture of ethanol and 2-propanol), then again sterilized in autoclave, and finally in Petri dishes inoculated with 0.1 ml of bacterial suspensions. Two densities of bacterial suspensions in physiological solutions were used, i.e. 0.5 and 1.0 of McFarland scale (1.5 ×10⁸ and 3.0 ×10⁸ CFU/ml). Incubation of bacteria on the top surfaces of samples was performed at 37 °C for 48 hours. Afterwards, bacteria were striped from tested surfaces using sterile swap and taken up in liquid culture medium for 48 hours. Finally, bacteria were pre-inoculated from liquid medium into the sodium chloride diagnostic soil in Petri dishes.

The anti-bacterial resistance of PBs in the diagnostic soil was assessed on the basis of the bacterial activity valued from 0 to higher numbers in CFU/ml.

**Mold test**

Resistance of the top surfaces of the PB’s samples (20 mm x 40 mm) to molds was performed with two microscopic fungi, the *Aspergillus niger* Tiegh. and the *Penicillium brevicompactum* Dierck. Samples were placed into sterilized Petri dishes φ 120 mm (two pieces per one dish filled with 3 – 4 mm thick layer of the 4.9 % Czapek-Dox agar, HiMedia
L. India) on plastic mats, and subsequently inoculated with a water spore suspension of the given mold species. Mold attacks of samples in thermostats lasted 28 days at 28 ± 2°C. The growth activity of moulds (GAM) on the top surfaces of samples was evaluated visually with help of a table magnifying glass by these criteria defined in the Standard STN 49 0604 (1980): 0 = no mould; 1 = mould up to 10 %; 2 = mould up to 25 %; 3 = mould up to 50 %, and 4 = mould more than 50 %.

Decay test
Resistance of the PB’s samples (40 mm × 40 mm × 7 mm) to decay was performed with the brown-rot fungus Coniophora puteana (Schumacher ex Fries) Karsten, BAM Ebw. 15. Samples were placed into sterilized Petri dishes ø 120 mm (one tested piece and one control piece per one dish filled with 3 – 4 mm thick layer of the 4.5 % malt agar, HiMedia Ltd., India) on plastic mats, in which a fungal mycelium was already grown up. The decay process lasted 6 weeks at 24 ± 2°C. The intensity of decay in the PB’s samples was evaluated on the basis of their mass losses (Δm). For these aims, the samples before and after decay tests were air-conditioned at a temperature of 20 ± 2°C and a relative humidity of 65 ± 2 % to achieve a constant weight (with an accuracy of 0.001 g). The mass losses of samples were determined from their weights in conditioned states before (m0) and after fungal attack (mFungal-Attack), by the Eq. 1.

\[ \Delta m = \frac{m_0 - m_{\text{Fungal-Attack}}}{m_0} \times 100 \ \ [%]\]

RESULTS AND DISCUSSION

Results related to increased biological resistance of the copper-modified PBs – containing copper sulphate pentahydrate (CuSO₄·5H₂O) – are present in Table 2.

Tab. 2 Biological resistance of the copper-modified particleboards to bacteria, molds, and the brown-rot fungus C. puteana.

<table>
<thead>
<tr>
<th>Particleboard</th>
<th>Bacterial activity (10⁶ CFU/ml)</th>
<th>Mold growth activity (0-4)</th>
<th>Mass loss (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CuSO₄·5H₂O (w/w in MUF glue)</td>
<td>Staphylococcus aureus</td>
<td>Escherichia coli</td>
<td>Penicillium brevicompactum</td>
</tr>
<tr>
<td></td>
<td>0.5*</td>
<td>1.0*</td>
<td>0.5*</td>
</tr>
<tr>
<td>0</td>
<td>0.17</td>
<td>0.33</td>
<td>0.18</td>
</tr>
<tr>
<td>2</td>
<td>0.18</td>
<td>0.28</td>
<td>0.12</td>
</tr>
<tr>
<td>6</td>
<td>0.16</td>
<td>0.31</td>
<td>0.15</td>
</tr>
<tr>
<td>12</td>
<td>0.09</td>
<td>0.17</td>
<td>0.13</td>
</tr>
<tr>
<td>24</td>
<td>0.04</td>
<td>0.07</td>
<td>0.09</td>
</tr>
</tbody>
</table>

Notes:
- Mean values of the bacterial activity are from 3 samples, of the mold growth activity from 6 samples, and of the decay activity of C. puteana from 6 samples.
- Standard deviations are in the parentheses.
- * Represents density of the bacterial suspension in the McFarland scale (0.5 or 1.0).

The anti-bacterial properties of the copper-modified PBs were higher comparing to control unmodified PBs. Activity of the gram-positive bacteria Staphylococcus aureus decreased from 0.33 up to 0.07 ×10⁸ CFU/ml (at using 1.0 McFarland bacterial inoculum) or from 0.17 up to 0.04 ×10⁸ CFU/ml (at using 0.5 McFarland bacterial inoculum) – it means maximally about ~77 %. Activity of the gram-negative bacteria Escherichia coli decreased
from 0.35 up to $1.5 \times 10^8$ CFU/ml (at using 1.0 McFarland BI) or from 0.18 up to $0.09 \times 10^8$ CFU/ml (at using 0.5 McFarland BI) – it means maximally about 57 or 50 %. FEYZI et al. (2017) found that bacterial organisms, such as S. aureus and E. coli, are susceptible to 6.25 µg/ml or lower concentrations of CuSO$_4$.5H$_2$O in water solutions.

The anti-mold efficacy of CuSO$_4$.5H$_2$O on the top surfaces of PBs at reducing growth of the molds Aspergillus niger and Penicillium brevicompactum (valued in scale 0 – 4) was a partly milder. Growth activity of P. brevicompactum decreased from 2.67 up to 2.0, i.e. maximally about 25.1 %, and of A. niger from 3.17 up to 2.17, i.e. maximally about 31.5 %. It is in consistence with results about a poor mold-resistance of commercial PBs (CHUNG et al. 1999, STANGIERSKA et al. 2013, VIDHOLDOVÁ et al. 2015), and a slightly higher mold-resistance of plywood with surfaces treated with CuSO$_4$.5H$_2$O (REINPRECHT et al. 1986).

The anti-decay efficacy of copper sulphate pentahydrate added to the PBs was significant against the brown-rot fungus Coniophora puteana, since mass losses of the copper-modified PBs decreased from 17.38 up to 9.64 %, i.e. maximally about 44.5 %. The linear correlation with a coefficient of determination $R^2 = 0.51$ confirmed that a higher amount of CuSO$_4$.5H$_2$O in MUF glues and following also in the copper-modified PBs was connected with suppression of decay in PBs determined on the basis of their lower mass losses (Figure 1). Examples from the growth intensity of C. puteana mycelia on surfaces of PBs are present in Figure 2.

![Fig. 1 Linear correlation between the increased amounts of CuSO$_4$.5H$_2$O in MUF glues (w/w %) used at preparation of PBs and the increased resistance of copper-modified PBs against rot by C. puteana determined through their mass losses (Δm %).](image)

Biological resistance of PBs depends mainly on these three factors: - applied biocide, - used wood particles and glues, - production technology. STANGIERSKA et al. (2013) by addition of organic fungicides into glues (a mixture of didecyldimethylammonium nitrate and 1,2,4-triazole derivative introduced into amino glues in amount of 3.0 kg per m$^3$ of PB) prepared PBs completely protected from degradation by the fungus C. puteana. SEN et al. (2010) tested different “wooden composite – cardboard” panels made from recycled carton with use of four glue types applied between the wood veneer and the cardboard, i.e. melamine-urea formaldehyde (MUF), polyurethane (PU), phenol-formaldehyde (PF), and urea-formaldehyde (UF) glue. They found that the most resistant against decay by the fungus C. puteana was composite prepared with the MUF glue, at which small mass losses by the Standard EN 113 (1996) determined as well as for other composites prepared with other glue types: $\Delta m = 0.43$ % (MUF), 0.47 % (PU), 0.69 % (PF), and 0.47 % (UF).
Fig. 2 Growth of the brown-rot fungus *Coniophora puteana* on surfaces of the copper-modified PBs (MUF glues treated with 2 to 24 w/w % of CuSO₄·5H₂O) and the control PBs after 6 weeks.

Generally, on the basis of experiments achieved in this and other works can be stated that special particleboards, cardboard or other wooden composites glued preferentially with MUF adhesives and if necessary with addition of ecologically-friendly biocides could be in some situations recommended as well as for use in more humidity conditions – some of them also for bathroom furniture or roofing materials which are exposed to fungal risk areas.
CONCLUSIONS

Copper sulphate pentahydrate added into melamine-urea formaldehyde glues in the amounts of 0, 2, 6, 12 or 24 w/w % increased selected biological properties of the 1-layer particleboards:

- a resistance to the gram-positive bacteria *Staphylococcus aureus* by up to 76.5 % and the gram-negative bacteria *Escherichia coli* up to 50.0 %,
- a resistance to the molds *Penicillium brevicompactum* and *Aspergillus niger* by up to 25.1 % and 31.5 %,
- a resistance to the brown-rot fungus *Coniophora puteana* by up to 44.5 %.

The copper-modified PBs with an increased biological resistance have potential to be for a short time exposed in partly wetted indoor environment, e.g. at accidently increased moisture. Of course, in such cases, there also are very important good moisture properties of PBs – small thickness swelling and water absorption (see IŽDINSKÝ and REINPRECHT 2017).

REFERENCES


ACKNOWLEDGEMENTS

This work was supported by the Slovak Research and Development Agency under the contract No. APVV-0200-12. We thanks also to J. Kapustová, V. Hyšková, M. Oros and J. Krokošová for their technical assistance.

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