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# Characterization of packaging ability to protect beer from light degradation and introduction of a new Packaging Riboflavin Index

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## Abstract

Bottled beer is exposed to light during transport or storage, which can unfavourably affect its sensory properties. One of the functions of the packaging is to provide maximum protection against possible light degradation. Riboflavin (vitamin B2), which acts as a photosensitizer, is a key substance for the occurrence of light degradation in beer. Riboflavin absorbs light in the visible region of the spectrum and transfers its energy to other compounds. A cascade of subsequent chemical reactions leads to sensory damage to the beer. Based on the transmission spectrum of packaging (glass and plastic), the Packaging Riboflavin Index (**$P_{RF}I$**)was introduced. **$P_{RF}I$** was designed as a tool which quantitatively assesses the ability of packaging to protect beer from light damage. The basic types of commercially used glass and plastic packaging were compared using this index. The validity of the index was verified in an experiment with defined light damage of beer in different packaging.

**Keywords:** beer; light degradation; riboflavin; skunky flavour; light-struck flavour; 3-methyl-2-butene-1-thiol

## 1 Introduction

When beer is exposed to light radiation, an off-flavour known as “light-struck” can develop. A description like skunky or monkey pavilion is used to characterize it. The substance 3-methyl-2-butene-1-thiol (MBT) is primarily responsible for this flavour.

The sensory perception of MBT is very intensive. The pure substance has a pungent odour, referred to as “skunk” due to its similarity to the odour of skunk excrement. Threshold values detectable by human smell range from 0.2 to 0.4 ng/l in water. The substance is slightly less sensory active in beer with a threshold detection 4–35 ng/l (Templar et al., 1995).

The formation of MBT is associated with non-enzymatic reactions of isohumulones with sulphur components of amino acids and their derivatives. The main route MBT formation involves the decomposition of isohumulones into 3-methylbut-2-enyl radical, while the source of thiol radicals is sulphur-containing proteins or amino acids. The decomposition of isohumulones is catalysed by photosensitive riboflavin (RF). The combination of both radicals finally results in MBT (Sakuma et al., 1991). RF was found to be necessary as a so-called photosensitizer for the formation of MBT from isohumulones and sulphur proteins because it absorbs light radiation and subsequently enables the transfer of absorbed energy to other substances (Heyerick, 2001).

The part of the light that passes through the packaging and can cause riboflavin excitation is dangerous for beer. Knowing the spectral transmittance T(λ) of the package and the absorption curve of riboflavin $A_{RF}(\lambda)$, it is possible to determine how much radiant (light) energy passes through the wall of the package and is absorbed by riboflavin. The introduction of an index that can characterize the potential ability of the packaging to protect beer from light damage would be greatly useful for practice.

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### *1.1 Glass bottle as a light barrier*

The packaging, with its properties, can contribute to a better or worse degree of protection of the beer against light radiation. Beer is perfectly protected only in completely opaque packaging, such as metal barrels or cans. However, light can pass through glass or plastic bottles to a certain extent and interact with the beer causing its sensory damage. The degree of damage is determined by the spectral composition of the incident light, the intensity of the light at the point of impact on the beer bottle and the spectral dependence of the transmittance of the packaging for potentially dangerous radiation (Gabriel et al., 2022).

Glass is made from a mixture of glass sands, which contain 60–80% silica, as well as limestone, sodium carbonate and potassium carbonate. Clear glass effectively absorbs UV radiation, which practically eliminates its destructive effect on beer. However, clear glass is transparent to radiation from the visible region. The amount of light falling on the beer can be reduced by appropriate colouring of the glass used. Due to the colouring, the light is absorbed by the material of the bottle and significantly reduces its intensity before entering the beer itself.

Coloured glasses can be obtained by adding small amounts of oxides that form coloured silicates. For example, adding CoO makes the glass blue and a larger amount of $FeSiO_3$ colours the glass black, while a small amount causes the deep green. Glass is particularly strongly coloured by $Fe_3O_4$ especially when compared to trivalent iron itself, which causes yellow-green to brownish-yellow hue. Manganese oxide with a large amount of $Fe_2O_3$ gives the glass a brown colour. Further, chromium, uranium and vanadium oxides also lead to green tint of the glass. The ruby red colour is due to $Cu_2O$ and colloidally dispersed gold, while the lighter red requires adding of Se and Cd. The yellow colouring can be reached by Ag, Ce and Ti. Historically, two colour designs have been established for glass beer bottles, i.e. green and brown. However, there are also other coloured bottles (e.g. blue) or bottles made of almost clear glass.

Even among the “classic” beer bottles of green or brown colour, there is a wide range of colour shades. The intensity of light that passes through the bottle wall depends on the specific absorption coefficient of the bottle material and also on the wall thickness in accordance with the Lambert-Beer law. The radiation intensity decreases exponentially with increasing bottle thickness. As the thickness of the bottle walls decreases, its weight decreases, but at the same time the intensity of the radiation that can get into the beer increases significantly. Differences in the ability of different glass bottles to protect beer from light damage have been known for a long time (Gamer et al., 1964; Taylor and Poole, 1971).

### *1.2 PET bottles as a light barrier*

Plastic bottles are made from a polymer material, semi-crystalline polyethylene terephthalate (PET). Production is based on two basic raw materials, ethylene glycol monomer and terephthalic acid or its ester. Bottles are produced in different shapes, volumes and can be offered in a wide range of colours using various additives and dyes (Christensen, 2003; Stowitts, 2015).

The protective function of the colour of PET bottles has not been paid much attention yet. In terms of shelf life and sensory changes of beer in PET bottles, the problems of gas diffusion through the polymeric wall of the bottles have been addressed so far. Specifically, the research focused on the gradual decrease of $CO_2$ in beer over time and the increase of oxygen content (Orzinski et al., 2005). The effect of increased oxygen concentration on the development of many undesirable flavours is well known. In addition, in cheaper simple PET bottle designs, oxygen diffusion rapidly reduces the sensory stability of beer and its overall shelf life (Bachvarov and Marinova, 2006; Profaizer, 2007). A higher oxygen level, on the other hand, effectively suppresses the formation of light-struck flavour, because oxygen is able to take energy from the excited state of riboflavin and prevent the generation of MBT. However, with increasing technological progress, PET bottles with active protective layers have appeared, i.e. barriers significantly reducing gas diffusion through the bottle wall, which should extend the shelf life of beer.

However, a missing and important aspect is the ability of the packages to protect beer from light damage (Boutroy et al., 2006; Cahill et al., 2002; Di Felice et al., 2008; Folz, 2010).

The aim of this work is to compare the ability of transparent packaging (glass and PET bottles) to protect beer from light damage. At the same time, we introduce an index that quantifies the potential the packaging has to protect beer from light damage and validate it experimentally on a set of commercial packaging.

## 2 Materials and methods

### *2.1 Material*

A riboflavin solution was prepared by dissolving 10 mg of riboflavin (for biochemistry, Sigma-Aldrich) in deionized water. Deionized water was prepared using an Aqual 35 instrument. The conductivity was less than 0.2 μS/cm. The absorption spectrum of riboflavin and the absorption spectra of glass and plastic bottles were measured on a single-beam spectrophotometer Specord 40 from Analytik Jena. Samples of 3 × 3 cm were cut out of the beer bottles for absorption measurement.

Pilsner type lager beer (alcohol 5.0% vol., original wort extract 12.45% wt., colour 9.8 EBC, turbidity 0.31 EBC) was used for the measurement. Beer was produced in an experimental brewery and bottled under a protective atmosphere into uncoloured clear, green and brown glass bottles corresponding to the standard type 0.5 l Nord-Rhein Westfalen (NRW) bottles. For technological reasons, it was not possible to pour the same beer into plastic bottles at the same time. The effect of the packaging colour was simulated by covering the outside of clear glass bottles with a layer of plastic from standard plastic bottles.

Samples of 0.5 l glass bottles were tested. The white and blue bottles were purchased from the glass beer bottle supplier Bricol GmbH (samples marked GC – clear, GB – blue). Samples of green and brown glass 0.5 l NRW type bottles from various Czech breweries were purchased at retail. The labels were removed from the bottles and they were washed. A total number of 5 types of green bottles (samples marked GG1 to GG5) and 3 brown bottles (marked GB1 to GB3) were tested. To test the plastic packaging, beer samples in 2 green (samples labelled PETG1 to PETG2) and 4 brown 1.5 l PET bottles (samples labelled PETB1 to PETB4) were purchased from a retail store. The bottles were emptied and washed.

### *2.2 Packaging Riboflavine Index ($P_{RF}I$ )*

We introduced the Packaging Riboflavin Index ($P_{RF}I$), which quantified the potential of a selected bottle to protect beer from light damage. We defined the index as the spectral fraction of the luminous flux that passed through the packaging and fell within the riboflavin absorption region. The index was calculated according to equation (1) as the integral of the product of the wall transmittance curve **$T(\lambda)$** of the given bottle with the riboflavin absorption coefficient **$A_{RF}$ $(\lambda)$** over wavelengths where neither of these values is zero. In the denominator is the integral of the riboflavin absorption coefficient, this means the same integral as in the numerator with a constant value of $T(\lambda)=1$ corresponding to the non-absorbing packaging. A value of 300 nm was chosen as the lower limit of the integration, because the transmittance of all common bottles is zero for shorter wavelengths. The wavelength of 780 nm was chosen as the upper limit, which is the limit of the visible light. Also 550 nm could be used as an upper limit because higher wavelengths are not absorbed by riboflavin and the absorption coefficient is zero.

$$O_{RF}I = \frac{100 * \int_{300}^{780} A_{RF}(\lambda) * T(\lambda) * d\lambda}{\int_{300}^{780} A_{RF}(\lambda) * d\lambda} \tag{1}$$

### *2.3 Colorturb apparatus – measurement of absorbance of samples in closed commercial bottles*

The Colorturb apparatus (Gabriel and Sigler, 2018) was used to evaluate optical damage caused by illumination of beer in closed bottles. The Colorturb apparatus is equipped with a measuring chamber filled with an immersion liquid and rotates the sample during the measurement period. This arrangement allows accurate measurement of optical signals in transmission and nephelometric mode directly in closed commercial glass bottles with relatively significant wall inhomogeneities. The apparatus uses a 3-color LED with central wavelengths of 466 nm, 522 nm, and 634 nm for the blue, green, and red regions as a light source. This enables an independent measurement of sample absorbance at these wavelengths. AbsBlue is the absorbance at 466 nm.

The effective optical path of the bottles was determined by a control measurement of the absorbance of the clear bottles used that were filled with a coloured solution. The absorbance of solution was previously measured on a spectrophotometer. The mean optical path of the measured bottles was 50 mm with a standard relative deviation of ± 0.6%. Due to this low deviation, it was not necessary to correct the optical signals for the different diameters of the individual bottles.

### *2.4 Optical method of determining the degree of light degradation of beer*

The measurement of changes in absorbance of the sample directly in the bottle was used to determine the light degradation degree of beer. The light degradation of beer is associated with the breakdown of riboflavin, which causes changes in absorbance at around 450 nm (Pozdrik, 2006; Gabriel et al., 2022). The decrease in absorbance corresponds to the loss of riboflavin due to its decomposition during the light degradation of beer. Riboflavin has a significant absorption peak at the wavelength of 450 nm, RF decomposition is associated with a decrease in sample absorbance in this region. The LSFS index introduced in previous article (Gabriel et al., 2022) was used as a measure of beer degradation. The LSFS index was calculated as a linear coefficient of a straight line interpolated by AbsBlue values in the initial phase of the degradation.

### *2.5 Experimental verification of the validity of the packaging index*

The validity of $P_{RF}I$ index was tested by the following experiment. One type of beer was bottled under a protective atmosphere into selected bottles with different spectral absorption of their walls. The bottles were illuminated in a defined manner (time, light intensity) and the degree of light degradation was evaluated. The measure-

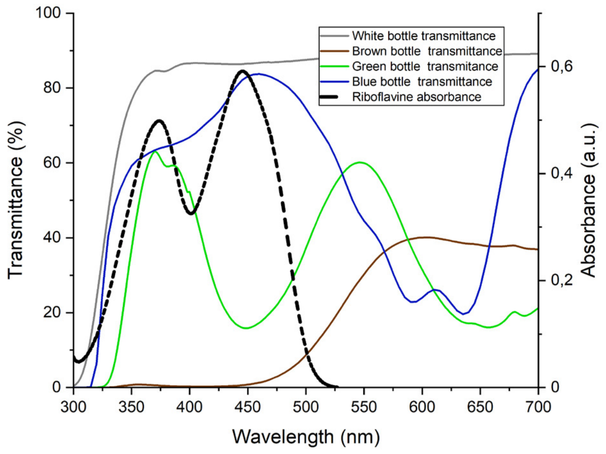

**Figure 1** *Comparison of RF absorption spectra and transmission spectra of green, brown, blue and clear beer glass bottles*

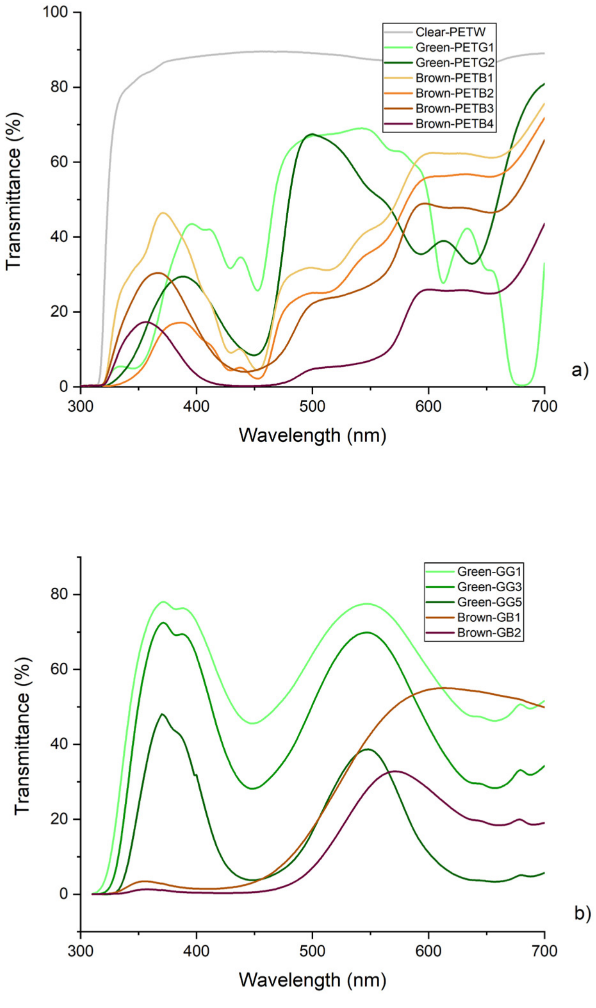

**Figure 2** *Transmission spectra of green and brown PET bottles a) and glass bottles b)*

ment of changes in absorbance of the AbsBlue sample directly in the bottle on the Colorturb was used to determine the degree of beer light degradation. To illuminate the samples, an LED bulb (1500 lm) with white light of the Coolday-light type, 6500K in a diffuse design was used as a standard light source. The light bulb was placed in a holder in a vertical position and the test beer bottles were placed around it at the same distance. The illumination intensity at the point of impact on the bottle walls was checked with a calibrated Digital Instruments LX1108 luxmeter and was the same for all samples.

The samples were illuminated for a defined period of time at regular intervals. Then they were placed in the Colorturb apparatus, where the AbsBlue value was measured. The LSFS index was calculated from the decrease in the AbsBlue value depending on the illumination time as a measure of the light degradation of the sample. Clear (GC), blue (GB), green (GG3) and brown (GB1) glass bottles and brown plastic bottles (PETB1 and PETB4) were selected for experimental verification of the $P_{RF}I$ index. Pieces were cut from the plastic bottles to cover the white NRW bottles filled with the same beer.

## 3 Results and discussion

### 3.1 Absorption spectrum of riboflavin

The absorption spectrum of riboflavin is shown in Figure 1. The spectrum was measured with a Specord spectrophotometer. Light with a wavelength shorter than 300 nm does not penetrate transparent packaging. The RF spectrum has a broad absorption band in the short-wavelength region of visible radiation between 300–500 nm with 2 peaks at 375 and 445 nm. Light with a wavelength longer than 510 nm is no longer absorbed by RF. Therefore, the most dangerous radiation for beer is the radiation that contains a short-wave component in the 300–500 nm region.

### 3.2 Spectral characteristics of glass and plastic bottles

The transmission spectra of selected glass and plastic beer bottles with different colours were measured on a Specord spectrophotometer. Figure 1 illustrates a comparison of the measured transmission spectra of glass green, brown, blue and clear beer bottles and the absorption spectrum of RF. Figure 2 shows the transmission spectra of the wall material of several types of green and brown glass and plastic beer bottles.

It is clear from Figure 1 that even clear uncoloured bottles transmit almost 90% of the incident light in the visible spectrum (10% of the light is reflected on the surfaces of the bottle), but they absorb all radiation below 300 nm very effectively, both in the glass and PET design (Figure 2). The undesirable beer flavour in both, glass and PET containers can be generated only by visible radiation, i.e. in other words only by that part of the radiation which is absorbed by riboflavin. Therefore the bottle should be of a colour that absorbs as much of this radiation (blue to blue-green light) inside its walls as possible. The walls of the brown bottle absorb blue and blue-green light and transmit light only in the red part of the spectrum with a wavelength longer than 500 nm, where RF almost no longer absorbs. The green bottle transmits light in the blue and green part of the spectrum in the areas of absorption peaks of RF at 450 nm and especially at 380 nm (Figure 1).

Therefore, beer in a green bottle is much more susceptible to light damage than that in a brown bottle. A blue bottle transmits most of the incident radiation from the spectral region of RF absorption between 300–500 nm (Figure 1). Its ability to protect the beer from light damage is minimal. Marketing efforts to differentiate the product, for example by packaging it in a blue bottle, are thus detrimental to its quality.

Brown glass bottles transmit only a percentage of light units (up to 5%) in the spectral region around 450 nm. The situation is different for green bottles (Figure 2b). There is much more variation between different types of bottles; some light green bottles will transmit even 45% light at 450 nm, while dark green bottles less than 10% light. Thus, the differences in transmittance are even fourfold. However, at 380 nm, the variation in transmittance is even greater, radiation from about 45% to 80% can pass through the green bottles. The transmission spectra of all measured glass bottles, both in the group of green and brown ones, have the same shape. Similar technological procedures and colouring additives are used in the production of coloured beer glass. The various colour shades are mainly caused by different wall thickness or the concentration of additives.

Transmission spectra (Figure 2a) of PET bottles are more complex. The spectra differ significantly from each other, whether it is a group of green or brown bottles. The more complicated shapes of transmission spectra result from a much wider range of additives and dyes for colouring plastic bottles. Brown PET bottles, like glass bottles, significantly suppress the radiation around 450 nm. However, the transmittance around 380 nm in some brown PET bottles is not negligible in contrast to brown glass bottles, the values can reach 10–40%.

### 3.3 *Packaging index of glass and plastic bottles*

The $P_{RF}I$ packaging index defined according to the equation 1 will allow a simple quantitative comparison of the protection degree provided by different types of beer bottles against the possible development of a light-struck/skunky flavour. A clear bottle that would transmit all radiation (T=1 in the entire spectral region) will have a $P_{RF}I$ index value of about 100 and provide no protection to the beer. If no critical radiation passes through the bottle (T = 0), the value of the $P_{RF}I = 0$. On the contrary, a double index value of one package compared to another means that the same damage will be caused to the beer in that package under the same lighting, but in half the time compared to the package with half the index value.

The $P_{RF}I$ packaging index values for the tested beer bottles were calculated according to equation 1. The values are shown in Table 1. The $P_{RF}I$ values of the brown bottles are generally lower than those of the green bottles. The brown bottles gave the $P_{RF}I$ values below 10 with the exception of PET bottles PETB1 (14.2) or PETB3 (23.8). Even so, this value is lower than the indices of green bot-

**Table 1** *Packaging RF index ($P_{RF}I$) for selected glass and PET beer*

| Sample | Bottle Colour | Bottle Material | Packaging RF index ($P_{RF}I$) | $P_{RF}I_{rel}$ (clear bottle/bottle) |
|---|---|---|---|---|
| GC | Clear | Glass | 82.6 | 1.0 |
| GB | Blue | Glass | 69.7 | 1.2 |
| GG1 | Green | Glass | 57.5 | 1.4 |
| GG3 | Green | Glass | 45.1 | 1.8 |
| GG5 | Green | Glass | 18.1 | 4.6 |
| GB1 | Brown | Glass | 3.6 | 22.9 |
| GB3 | Brown | Glass | 1.0 | 82.6 |
| PETG1 | Green | PET | 32.5 | 2.5 |
| PETG2 | Green | PET | 20.5 | 4.0 |
| PETB1 | Brown | PET | 23.8 | 3.4 |
| PETB2 | Brown | PET | 9.8 | 8.4 |
| PETB3 | Brown | PET | 14.2 | 5.8 |
| PETB4 | Brown | PET | 7.7 | 10.7 |
| PETC | Clear | PET | 85.5 | 0.97 |

tles, where only the darkest green bottles achieved comparable values (18.1 for the glass bottle GG5 or 20.5 for the plastic one PETG2).

A clear glass bottle provides minimal protection to beer as it only absorbs radiation with a wavelength shorter than 350 nm. If we accept this bottle as the least suitable packaging in terms of its ability to protect beer from destructive light, we can compare it to other bottles. To compare the packaging index of individual samples with a clear bottle, the $P_{RF}I_{rel}$ value was introduced as the ratio of the packaging index of the clear glass bottle (GC) to the index of the tested bottle (XX): $P_{RF}I_{rel} = P_{RF}I\ (GC) / P_{RF}I\ (XX)$.

$P_{RF}I_{rel}$ values are given in the last column in Table 1. $P_{RF}I_{rel}$ value indicates how many times a given bottle provides better protection to beer than the clear bottle, or how many times longer the beer in the selected bottle must be exposed to light to cause the same light degradation as in a clear bottle. The $P_{RF}I_{rel}$ values allow a quick and practical comparison of bottles.

Table 1 demonstrates that the blue bottle (GB) is only 20% more effective in protecting beer from light than the clear bottle (GW). There are orders of magnitude differences even between commonly used bottles. The brown glass bottle GB3 provided 83× better protection than the clear bottle and 59× better protection than the green glass bottle GG1. Therefore, under identical lighting conditions, the same degradation should occur to the beer in the brown bottle GB3 in 83× longer time interval than in the green bottle GG1.

A comparison of glass and PET packaging indicates that PET bottles surprisingly provide comparable or even better protection to beer compared to glass in the case of the green bottle. On the contrary, glass absorbs light, i.e. it protects the beer significantly more than PET packaging in the case of the brown packaging. Brown bottles generally protect the beer from the development of light-struck/skunky flavour substantially better than green bottles. Nevertheless, e.g. a dark green glass bottle (GG5) displayed at least comparable or even better results than a light brown PET bottle (PETB1).

The effect of the spectral characteristics of the used light source, as well as the intensity of the incident radiation on the sample and its dose was neglected when introducing the packaging index according to equation 1. The reason for this simplification was the requirement that the introduced index depends only on the properties of the bottle, not on the properties of the light. The influence of the spectral characteristics of the light source on beer damage will be the subject of another research.

### *3.4 Experimental verification of $P_{RF}I$ index*

An experiment with the defined illumination of beer in a selected bottle was performed to verify the packaging index. Figure 3 illustrates the absorbance (AbsBlu) curves at 466 nm measured on COLORTURB as a function of illumination time. The LSFS index was calculated from the decrease in absorbance (Gabriel et al., 2022). Nevertheless, AbsBlue decrease in sample GB1, i.e. brown glass bottle with an **$P_{RF}I$** index of 3.6, was unmeasurable (less than the measurement error) and therefore the LSFS index could not be determined. This case is not reported in the results.

Table 2 demonstrates the values of the **$P_{RF}I$** index calculated according to equation 1 from the transmission spectra of the investigated bottles. Column 2 gives the values of the **$P_{RF}I_{rel}$** index relative to a clear uncoloured glass (GC) bottle. At the same time, the **LSFS** index values characterizing the degree of light degradation of the samples are shown in the table. **$LSFS_{rel}$** values are relative **LSFS** values referenced to the GC bottle.

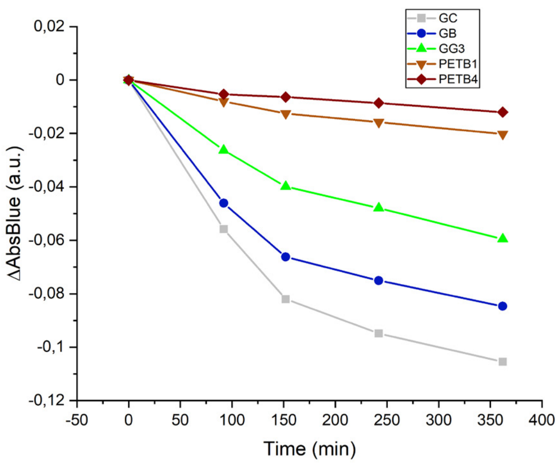

**Figure 3** *AbsBlue changes measured by the COLORTURB apparatus for beer in bottles with different spectral transmittance when illuminated by a CoolDayLight LED lamp, 6500 K*

**Table 2** *Values of the $P_{RF}I$ index and the degree of optical damage LSFS and their relative ratios related to a clear glass bottle*

| Bottle | $P_{RF}I$ | $P_{RF}I_{rel}$ | LSFS × $10^{-5}$ ($min^{-1}$) | $LSFS_{rel}$ |
|---|---|---|---|---|
| Clear glass – GC | 82.6 | **1.0** | 60.6 ± 1.0 | **1.0 ± 0.1** |
| Blue glass – GB | 69.7 | **1.2** | 50.1 ± 1.0 | **1.2 ± 0.1** |
| Green glass – GG3 | 45.1 | **1.8** | 28.6 ± 1.0 | **2.1 ± 0.3** |
| Brown PET – PETB1 | 23.8 | **3.5** | 8.7±1.0 | **6.8 ± 1.0** |
| Brown PET – PETB4 | 7.7 | **10.7** | 5.8 ± 1.0 | **10.5 ± 2.0** |

The calculated $\mathbf{P_{RF}I_{rel}}$ values corresponded to the light degradation of the samples ($\mathbf{LSFS_{rel}}$). The only exception was the light brown plastic bottle (PETB1), where the damage of the beer in the bottle was almost 2× lower than the calculated packaging index. This could be due to imperfect contact of the outer plastic packaging with the clear glass beer bottle. Imperfect contact could lead to uncontrollable light reflection at the plastic-glass transition. In case of brown bottles, the $\mathbf{LSFS_{rel}}$ value is burdened with a large error, because light degradation of beer and the corresponding decrease in AbsBlue are small. The obtained results confirmed the eligibility of the definition of the $\mathbf{P_{RF}I}$ packaging index.

## 4 Conclusion

The packaging RF index ($P_{RF}I$) was defined and calculated from the absorption spectrum of RF and spectral transmittance of the bottle wall. $P_{RF}I$ characterizes the bottle in terms of its potential ability to protect the beer from the light degradation. The transmission spectra of commonly used glass and plastic containers were measured and the $P_{RF}I$ index values were calculated. The validity of the packaging index definition was demonstrated experimentally by correlating the calculated index values with the rate of optical degradation of beer in different packaging. The comparison of the $P_{RF}I$ indexes calculated for selected types of commercially used bottles showed that there are orders of magnitude differences between them. The comparison of glass and PET packaging demonstrated that green PET bottles provide comparable or even better protection for beer than green glass bottles, while brown glass packaging absorbs light (protects beer) significantly better than PET. Brown bottles generally protect beer significantly better than green bottles.

## 5. Acknowledgement

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