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Butylated hydroxytoluene (BHT) – Evaluation of a BAR

Assessment Values in Biological Material – Translation of the German version from 2023

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Abstract

In 2012, the German Commission for the Investigation of Health Hazards of Chemical Compounds in the Work Area derived a maximum workplace concentration (MAK value) for butylated hydroxytoluene (BHT) [128-37-0] of 10 mg/m^3 (inhalable fraction). Measuring body burden after exposure to BHT showed BHT acid (3,5-di-tert-butyl-4-hydroxybenzoic acid) in urine to be an adequate biomarker. To date, no data have been published on urinary concentrations of BHT acid after occupational exposure to BHT with correlation to air concentrations of BHT or with effects from BHT exposure; as a result, no biological tolerance value (BAT value) can be derived. There are, however, data available for the derivation of a biological reference value (BAR). Based on the available data, a BAR of 7 µg BHT acid (after hydrolysis)/l urine has been established. Sampling time is at the end of exposure or the end of the shift.

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BAR (2021)	**7 µg BHT acid (after hydrolysis)/l urine** Sampling time: end of exposure or end of the shift
MAK value (2012)	**10 mg/m³ I**
Absorption through the skin	–
Carcinogenicity (2004)	Category 4
Synonyms	2,6-Di-tert-butyl-p-cresol 2,6-Di-tert-butyl-4-methylphenol 3,5-Di-tert-butyl-4-hydroxytoluene
CAS number	128-37-0
Formula	$C_{15}H_{24}O$
Molar mass	220.35 g/mol
Melting point	70 °C (NCBI 2023)
Boiling point	265 °C (NCBI 2023)
Density at 20 °C	1.05 g/cm³ (NCBI 2023)

After its patent registration in 1947, butylated hydroxytoluene (BHT) was first used as a stabilising agent in the petroleum and adhesive industries. As early as the 1950s, its range of applications was expanded to include the stabilisation of food products and cosmetics due to its antioxidant properties (Nieva-Echevarría et al. 2015; Witschi et al. 1989). Its spectrum of uses is equally diverse in the food industry, where it is added to baking mixes, nuts, instant soups, chewing gums, fats, and oils. The maximum allowable added quantities are stipulated in the Regulation on Food Additives (Zusatzstoff-Zulassungsverordnung). For chewing gum, for example, this value is 400 mg/kg. In the List of Food Additives, this substance is labelled as E 321 (antioxidants) (EFSA 2012; Gries et al. 2020).

Further areas of application include pharmaceuticals (creams, gelatine capsules), cosmetics (shower gels, soaps), plastic and rubber products, as well as paints and lacquers. In 2000, its worldwide production volume amounted to about 62 000 t/a (OECD 2002).

BHT is marketed under various brand names, such as Vulkanox-BHT, Ionol, Paranox 441, Impruvol, or Antioxidant 4K.

Metabolism and toxicokinetics

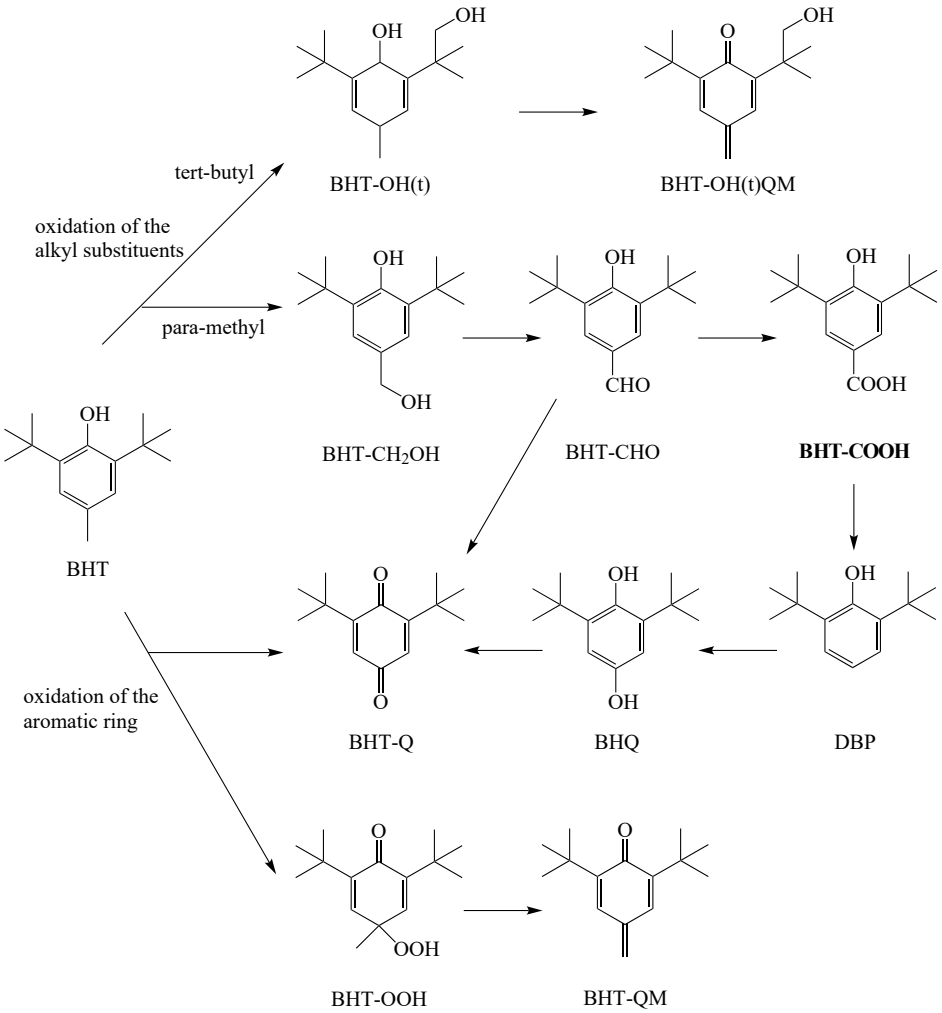
Absorption and distribution

The main absorption route of BHT in the workplace is inhalation. BHT may also be incorporated via the skin, whereby the systemically available amount of about 4% can be deemed minimal. BHT is rapidly resorbed from the gastrointestinal tract. After uptake, the substance is distributed in the liver and fatty tissues. The substance is predominantly excreted with the urine and faeces. Significant differences in types of metabolites and routes of excretion arise between species (Hartwig 2012).

Metabolism

Rats excreted between 80 and 90% of the orally administered dose with the urine within 4 days (Daniel and Gage 1965). In humans, 50% of the administered dose was excreted within one day and 66% within 11 days (Daniel et al. 1967).

Metabolism studies in rats, mice, and rabbits have shown that the formation of the identified metabolites is species-dependent. Metabolism can take place at three positions: ring, methyl group, and tert-butyl group. After oral administration, liver changes in rats and lung changes in mice were observed; these effects have been attributed to the formation of quinoid metabolites. It is not yet known whether quinone metabolites form in humans (EFSA 2012). An important BHT metabolite in humans is BHT acid (3,5-di-tert-butyl-4-hydroxybenzoic acid) (Nieva-Echevarría et al. 2015). Figure 1 provides an overview of the possible metabolites of BHT.



BHQ: 2,6-Di-tert-butyl-1,4-benzenediol; BHT: 3,5-Di-tert-butyl-4-hydroxytoluene; BHT-CHO: 3,5-Di-tert-butyl-4-hydroxybenzaldehyde; BHT-CH_2OH: 3,5-Di-tert-butyl-4-hydroxybenzyl alcohol; BHT-COOH: 3,5-Di-tert-butyl-4-hydroxybenzoic acid (BHT acid); BHT-OH(t): 2-Tert-butyl-6-(1-hydroxy-2-methylpropan-2-yl)-4-methylphenol; BHT-OH(t)QM: 2-Tert-butyl-6-(2-hydroxy-tert-butyl)-4-methylene-2,5-cyclohexadien-1-one; BHT-OOH: 2,6-Di-tert-butyl-4-methyl-4-hydroperoxy-2,5-cyclohexadien-1-one; BHT-Q: 2,6-Di-tert-butyl-2,5-cyclohexadien-1,4-dione; BHT-QM: 2,6-Di-tert-butyl-4-methylene-2,5-cyclohexadien-1-one; DBP: 2,6-Di-tert-butyl-4-phenol

Fig. 1 Overview of possible metabolites of BHT (according to Nieva-Echevarría et al. 2015)

Excretion

From the few available data on the metabolism of BHT in humans, it is known that up to 5% of the administered BHT dose is excreted as BHT acid with the urine and is primarily present as glucuronide (Nieva-Echevarría et al. 2015).

Critical toxicity

Independent of the route of exposure, BHT displays low acute toxicity. The majority of LD_{50} values are > 2000 mg/kg body weight. BHT has a slightly irritant effect on the skin and eyes of rabbits. Comprehensive data on critical toxicity can be found in the corresponding MAK Documentation (translated in Greim 2007). Systemic hepatic effects regarding tumour promotion are of utmost importance for the derivation of the MAK value.

The NOAEL (no observed adverse effect level) has been indicated as 25 mg/kg body weight and day and the ADI (acceptable daily intake) as 0.25 mg/kg body weight and day (EFSA 2012).

Exposure and effects

To date, no studies are available on the relationship between external and internal exposure or on the relationship between internal exposure and effects in humans.

Selection of the indicators

The specific urinary BHT metabolite BHT acid serves as an indicator for BHT exposure (Gries et al. 2020).

Analytical methods

After adding the $^{13}C_6$-ring-labelled internal standard, the samples are enzymatically hydrolysed in order to release the glucuronide-bound BHT-acid components. Via two-dimensional LC coupling, the analytical samples are, after online purification and enrichment, separated from matrix components by liquid chromatography and analysed by downstream tandem mass spectrometry. A quantitation limit (LOQ) of 0.2 BHT acid/l urine is hereby achieved (Gries et al. 2020).

Background exposure

In Germany, there have been several investigations on BHT exposure in the general population.

Göen et al. (2006) investigated 22 persons (7 women, 15 men) not occupationally exposed to BHT. In 91% of the samples, BHT-acid concentrations were measured in a range above the LOQ up to 12.7 µg/l urine.

In a collective of non-occupationally exposed persons, 72 of 80 investigated samples exhibited BHT acid values in a range above the LOQ up to 7.55 µg/l urine (Leng and Gries 2017). In this investigation, the 90th percentile was 1.89 µg BHT acid/l urine.

BHT acid was determined in the urine of 329 students between the ages of 20 and 29 years (Schmidtkunz et al. 2020). The 24-hour urine collection samples from the German Environmental Specimen Bank were taken in years 2000, 2004, 2008, 2012, 2015, and 2018. BHT acid was found in 98% of the samples. The median was 1.06 µg/l, the 95th percentile was 5.44 µg/l (Schmidtkunz 2022), and the maximum value was 18.1 µg BHT acid/l in 24-hour urine.

In their study, Wang and Kannan (2019) summarised data on background exposure to BHT. As large country-specific differences were evident (Japan: mean 5.49 µg BHT acid/l urine (median 3.86; range < LOQ–24.4); India: mean 5.15 µg/l (median 2.24; range < LOQ–24.0); China: mean 0.56 µg/l (median 0.26; range < LOQ–4.98); Saudi Arabia: mean 1.46 µg/l (median 0.44; range 0.11–5.64); USA: mean 7.44 µg/l (median 1.78; range < LOQ–46.0)) and as no 95th percentile is indicated in this study, these data will not be considered for the derivation of the BAR.

As part of the GerES V (German Environmental Study on the Health of Children and Youth 2014–2017), urinary concentrations of BHT acid were determined for 2091 persons aged 3 to 17 years. The median was 2.2 µg/l, the 95th percentile was 11.2 µg/l, and the maximum value was 248 µg BHT acid/l urine (Murawski et al. 2021).

Table 1 presents data on the background exposure of the general population to BHT.

Tab. 1 Urinary BHT-acid excretion of the non-occupationally exposed general population in Germany

Age	Sample size (% > LOQ)	BHT acid in urine [µg/l]			References
		Mean (median)	Range	95th Percentile	
Adults					
	22 (91%)	n. a. (0.91)	< LOQ–12.7	n. a.	Göen et al. 2006
	80 (90%)	1.05 (0.66)	< LOQ–7.55	*1.89*[a)]	Leng and Gries 2017
	329 (98%)	1.70 (1.06)	< LOQ–18.1	*3.28*[a)] 5.44[b)] 24-h urine collection	Schmidtkunz et al. 2020
Children and adolescents					
3–17 years	2091 (99.7%)	4.41 (2.18)	< LOQ–248	11.2	Murawski et al. 2021
3–5 years	337	7.04 (2.54)	< LOQ–248	23.1	
6–10 years	690	4.77 (2.40)	Maximum 152	13.6	
11–13 years	435	4.06 (2.22)	< LOQ–185	10.9	
14–17 years	629	2.84 (1.71)	< LOQ–72.5	7.20	

[a)] 90th percentile
[b)] Schmidtkunz 2022
LOQ: limit of quantitation; n. a.: not available

Evaluation

From the present data on background exposure to BHT based on the studies by

- Schmidtkunz et al. (2020) (95th percentile: 5.44 µg/l in 24-hour urine (Schmidtkunz 2022)),
- Göen et al. (2006) (maximum 12.7 µg/l urine), and
- Leng and Gries (2017) (90th percentile: 1.89 µg/l (range up to 7.55 µg/l))

a BAR of 7 µg BHT acid (after hydrolysis)/l urine

is established for BHT. Sampling should be carried out at the end of exposure or at the end of the shift.

The study by Murawski et al. (2021) concerns a collective of children and adolescents aged 3 to 17 years. The 95th percentile of 7.20 µg/BHT acid/l urine for the age group of 14 to 17-year-olds supports the BAR.

In a collective of 622 occupationally exposed persons, a 95th percentile of 9.71 µg BHT acid/l urine was found (Schmidtkunz 2022; Schmidtkunz et al. 2020). This collective cannot be used for the derivation of the BAR. It does show, however, that exposure levels from routine contact with BHT are of the same order of magnitude as the background exposure for the general population.

Interpretation

The BAR refers to normally concentrated urine in which creatinine content should be in the range of 0.3 to 3 g/l. In cases of urine samples outside the above limits, it is generally recommended to repeat analysis with a normally hydrated test person (translated in Bader et al. 2016).

Notes

Competing interests

The established rules and measures of the Commission to avoid conflicts of interest (www.dfg.de/mak/conflicts_interest) ensure that the content and conclusions of the publication are strictly science-based.

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