40 Years of total resin extract – Useful information from A to X (Part 2)

HOP EXTRACTION | In addition to pellets and CO_2 extracts, total resin extract (formerly referred to as ethanol extract) is a well-established hop product that has been utilized for decades in the production of beer. In the second article of this two-part series, the properties of total resin extract are presented in alphabetical order and, where applicable, the latest research findings are included as well.

IN THE FIRST INSTALLMENT (BRAUWELT International no. 1, 2022, pp. 12–15), keywords were defined, starting with A as in alcohol to H as in hard resins. Part 2 continues in this vein, beginning at I for iso-α-acids and continuing alphabetically to X for xanthohumol.

Iso-α-acids

During the production of total resin extract, a small portion of the α-acids derived from the hops become isomerized. When it was launched on the market, the iso-α-acid content was still around 3–4 %. After optimizing the process, it was reported in 1994 that the extraction was carried out under significantly milder conditions [1]. Since then, the iso-α-acid content of total resin extract has only amounted to around 1 %, which must be taken into consideration when producing beer. For this reason, total resin extract is analyzed using either the "total HPLC" method or the "LCBV" method.

- Total HPLC: iso-α-acids and α-acids (Analytica EBC method 7.8);
- lead conductance bitter value (LCBV): lead conductance value (LCV, Analytica-EBC method 7.6) + ½ iso-α-acids (Analytica EBC method 7.8).

In contrast to α-acids, only 50 percent of the iso-α-acids are accounted for in the lead conductance value. The other half must therefore be measured using HPLC (high pressure liquid chromatography).

Jubilee

Total resin extract had its 40th anniversary last year in 2021. Large-scale production began in 1981 (a time when there were no cell phones, and people were just beginning to purchase their first personal computers).

Kettle addition

In the brewery, the hop extract is usually added at the beginning of the boil in g alpha/hl (either according to total HPLC/hl or LCBV/hl). Total resin extract tends to dissolve better and isomerize more rapidly than the slightly less polar CO_2 extract. This difference can clearly be observed at both the laboratory and pilot scale. For example, in a trial conducted at a 120-liter pilot brewery, the yield of hop bitter substances was lower using CO_2 extract [7]. In the finished beer, this difference, compared to the total resin extract, was between 4 % and 7 %, depending upon the analysis method. However, the results of this research explicitly mention that "it may be assumed that, on an industrial scale, the

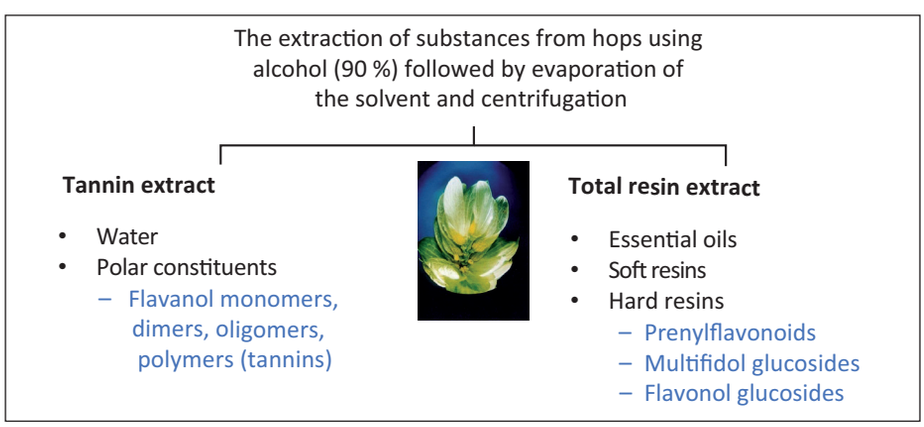

Fig. 1 Distribution of polyphenol hop compounds after extraction with alcohol

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difference in yield between ethanol and CO_2 extract will be lower because, on a pilot scale, the unfavourable ratio between wort volume and vessel surface area has a more pronounced effect".

This assessment was subsequently confirmed in the large-scale brewing trials mentioned above, in which no significant differences in yield could be determined [8].

The higher polarity of total resin extract is nevertheless a significant attribute that makes its use in brewing beer worthy of consideration, especially given that the duration of the boil in many breweries is now generally shorter and is carried out at lower temperatures.

Linalool and other polar hop aroma compounds

Since the process has been modified to create optimal conditions for the extraction process [1], almost all of the linalool and other polar hop aroma compounds (terpene alcohols, esters, ketones, epoxides) are nearly completely extracted from the hops. They can therefore be found in the total resin extract (> 90 %).

Myrcene and other terpenes

Losses of up to 50 percent remain unavoidable for myrcene and other monoterpenes, which all have low boiling points. By contrast, the rates of recovery in total resin extract are around 90 percent for the sesquiterpenes (humulene, caryophyllene) since their boiling points are higher [1].

Nitrate

Although the nitrate contained in the hops is extracted, the majority of it ends up in the aqueous phase of the tannin extract. As a result, total resin extract contains up to 98 percent less nitrate than was present in the whole hops prior to extraction [2].

Ochratoxin and other mycotoxins

Although mycotoxins may be present in other raw materials, such as barley and malt, hops and hop products are completely free of mycotoxins and thus they are not present in the total resin extract [3].

Dried whole hops and, above all, pellets and extracts in airtight packages create conditions which are extremely inhospitable for the growth of spores. These include a low residual moisture content and an anaerobic environment along with high concentrations of antimicrobial bitter substances.

Polyphenols

Fig. 1 shows the distribution of different kinds of polyphenols derived from hops after extraction with alcohol.

Together with the purely lipophilic components of the essential oils and the soft resin fraction, semi-polar polyphenols derived from the hard resin are present in the total resin extract.

Like all other water-soluble hop constituents (low molecular weight hydrocarbons and proteins, mineral salts), polyphenols with a greater degree of polarity are completely separated and end up in the aqueous phase with the tannin extract. This polyphenol fraction consists mainly of flavanols and, starting from the two monomeric compounds catechin and epicatechin, includes their dimers, oligomers and high molecular weight polymers (also called tanning agents or tannins).

Once the moisture evaporates to around 80 % dry matter, the tannin extract is sometimes blended with total resin extract at a specific ratio in order to obtain a defined (constant) α-acid content. This kind of "standard-

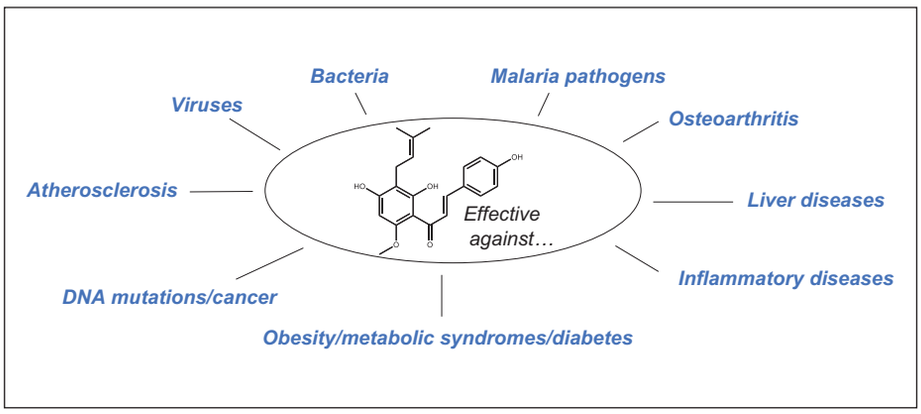

Fig. 2 Positive pharmacological properties of xanthohumol as reported in the scientific literature

ized extract" (e.g., 40% α-acids) was still quite popular in the early days of alcohol extraction but has since ceased to be relevant. It is now possible to obtain a specific α-acid content per unit of packaging (can) by precisely weighing the extract according to its α-acid content, which is determined analytically prior to filling and packaging.

Quality

After the total resin extract product was launched on the market in the early 1980s, numerous brewing analyses were conducted at the two renowned Chairs for Brewing led by Professors Narziß and Donhauser at the Technical University of Munich-Weihenstephan as well as at the Versuchsstation Schweizerischer Brauereien (VSB) headed by Dr. Pfenninger. All of them came without reservation to the conclusion that total resin extract is well suited for the production of high quality beers [4–6].

Brewing trials followed both on a pilot scale [7] and on an industrial scale, for example at the Diebels brewery [8]. Total resin extract and CO_2 extract were compared directly in these trials. As far as the quality of the beer was concerned, the results showed that they were virtually the same or the differences were imperceptible:

- All said, both extracts yield beers that hardly differ from one another in analytical and sensory terms [7];
- the beers could not be distinguished one from the other as far as the usual analysis characteristics are concerned nor indeed in tasting [8].

Recently, brewing trials comparing total resin extract and CO_2 extract were conducted once again and reported in the literature, but this time in a systematic comparison with the addition of hop pellets [9]. These studies, carried out at the Bitburger brewery, were presented for the first time at the EBC Congress in 2019 [10] and even received an award as the best poster presentation. The conclusion of this research is clear: it's all about the ratio! This study has shown that hop-accompanying bitter compounds can positively influence the quality of beer bitterness. Due to production conditions, CO_2 extract contains fewer phenolic compounds than ethanol extract. The addition of aroma hop pellets can partially compensate for this deficit. To achieve the same positive effect in CO_2 extract as in ethanol extract, more pellets must be added.

The aforementioned findings from the trials on the importance of the polyphenols in the hard resin have been corroborated in brewery practice. Due to their occurrence in pellets and total resin extract (i.e., ethanol extract), these products can be better substituted for one another.

Residues of pesticides

Hops used in the production of the extract must always conform to the upper limits for commercial hop products and comply with the Regulation on Maxiumum Residue Levels. The amounts of pesticides detectable in hops can be reduced even further during the extraction process. As various publications have shown, this largely depends upon the polarity of the substances.

The most recent publications on this topic present results comparing the recovery rates for all residues detectable in the total resin and CO_2 extracts produced from hops [11, 12]. Although there were specific differences attributable to the extraction method, neither kind of extract was more favorable in this regard over another. The results confirmed once again that the transfer rates of the individual chemical compounds largely depend upon their polarity.

Current research as part of a comprehensive annual program for monitoring these processes underscored these results while also taking the most recently approved chemical agents into consideration.

Spent hops

After the extraction, the spent hops are dried, then pelletized and sold for use in the production of animal feed. The alcohol recovered through condensation and rectification during the drying process is returned to be utilized again in the extraction cycle.

Milestones in research on xanthohumol (XN) and isoxanthohumol (IX)

1913	British scientists discover XN in hops
1961	Determination of the correct chemical structure of XN by Dutch scientists
1999	Publication on the behavior of XN during beer production: isomerization to IX (Oregon State University, USA)
1999	First reports on "in vitro" XN activity in terms of cancer prevention (Oregon State University, USA)
2002	Confirmation of the activity exhibited by XN by the German Cancer Research Center
2005	First reports on the anti-diabetic effects of XN in animal experiments (Japanese researchers)
2012	First reports on the prevention of mutations in DNA by XN in a human clinical trial (University of Vienna)
2020	First reports on the anti-diabetic and anti-inflammatory effects of IX in animal experiments (Japanese researchers)
2021	A daily allowance of 24 mg of XN per person is confirmed as safe by the FDA (US Food and Drug Administration) protocol. A clinical study is launched to investigate the role of XN in the prevention and treatment of inflammatory bowel disease (University of Portland and Oregon State University, USA)

Throughput of alcohol extraction plant

The Hallertauer Hopfenveredelungsgesellschaft in Mainburg extracts around 2000 kg of dried whole hops per hour. At three shifts per day (24 h), its daily capacity is just below 50 tons or approximately 250 tons per five-day work week. Depending upon the hop variety, this results in around 25–75 tons of total resin extract on a weekly basis.

Unique in the world

Since the process was patented and facilities at other locations have found it difficult to gain approval for alcohol extraction, the Hallertauer Hopfenveredelungsgesellschaft in Mainburg has remained the only place in the world where total resin extract is produced.

Viscosity

Due to its more complex composition and broader spectrum of bitter substances (including the hard resins), total resin extract is somewhat more viscous than CO_2 extract, although its viscosity varies as a function of the pressure conditions present during extraction. Generally speaking, the viscosity of total resin extract at 50 °C and that of CO_2 extract (supercritical, up to 300 bar) at 40 °C are comparable [1].

Water steam distillation

The final stage of the extraction process involves steam distillation in order to completely remove the alcohol. This entails subjecting the hops to vacuum evaporation at a maximum temperature of 60 °C. Since the high content of bitter acids means that the environment is slightly acidic and therefore has a pH of 5–6, similar to that of wort, the chemical reactions occurring at this stage are identical to those during the boil in the brewhouse. Consequently, a partial isomerization of the extracted α-acids takes place under these conditions. However, this is conducted in a vacuum and therefore at a lower temperature, which ensures that isomerization is kept to a minimum [1].

This final stage of the evaporation process facilitates the next step which is phase separation by centrifugation. This results in the complete separation of all of the extracted polar hop constituents (tannin extract). A type of pure resin extract is created at the same time, which contains the semi-polar polyphenols found in the hard resin but no water-soluble compounds whatsoever.

The steam distillation stage of the process was also the primary focus of the patent protection granted 40 years ago for the production of total resin extract.

Xanthohumol

In recent years, the body of knowledge regarding the significance of the hard resins in brewing has greatly increased. Research has also revealed the value of these substances for application in the field of pharmacology. The health-promoting (antioxidative) properties of quercetin have been known for a long time, and quercetin is not only found in hops but is very widespread throughout the plant kingdom. And yet, papers have appeared over the past few years in the scientific literature about prenylflavonoids, especially xanthohumol. These substances are unique to hops.

There are now more than 500 scientific publications discussing the promising attributes of this single compound derived from hops – starting with "in vitro" experi-

ments in test tubes through to "in vivo" experiments in animals to the first clinical studies in humans. The info box provides an overview of the historical development and the state of the research into the properties of xanthohumol (and isoxanthohumol).

As early as 2005, a complete issue of the well-known journal "Molecular Nutrition and Food Research" was dedicated solely to this substance derived from hops. The headline of the editorial was in the form of a question: "Xanthohumol – A New All-Rounder?" [13]. The astonishing variety of propitious characteristics has now been confirmed and is constantly being updated (fig. 2). Furthermore, all of these results also help to promote a positive image for hops and beer.

Currently, xanthohumol has even been considered as a means for treating SARS-CoV-2 infections [14]. Its efficacy and that of isoxanthohumol against the mechanism by which coronaviruses attack human cells are under investigation [15]. Both compounds are introduced to the brewing process through additions of total resin extract.

Summary

In the first few years after it was launched on the market, a number of technical improvements were carried out to optimize the production of total resin extract, and it has since become a valued hop product, one that is capable of meeting the current requirements of the brewing industry.

As a number of published brewing trials have shown, additions of pellets or CO_2 extract can be partially or completely substituted by total resin extract without any significant fluctuations in beer quality.

Although total resin extract has been on the market for a long time, the product's popularity has been buoyed by its natural composition and also by promising results from current research – both in terms of its value for the brewing industry and more generally for its positive effects on health. ■

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