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A Comparison of the Analytical and Brewing Characteristics of Cascade and Comet Hop Varieties as Grown in Yakima (USA) and Hallertau (Germany)

Hop varieties were and are transferred from one growing region to another. It is the great success of Cascade, the "lead variety" for dry hopping, that brought it to Germany. It is therefore of interest to discover whether there are differences between the Cascade hop grown in Yakima and that grown in the Hallertau.
To this end, a US Cascade (US-CA) was compared with a Hallertau Cascade (HCA) each from the 2012 and 2013 crops. The 2013 crop also provided a Comet from the USA (US-CM) and from the Hallertau (HCM). The hop samples were closely examined for aroma composition and polyphenols.
Systematic differences were found in both varieties with regard total and low-molecular polyphenols (Hallertau 50-78% higher). All three US hops have higher linalool contents. There are clearly higher quantities of the esters isobutyl-isobutyrate and 2-methylbutyl-2-methylpropanoate in the HCA of both crops. The differences in the other aroma components can be considered low.
Intensively hopped beers were brewed on a 2hl scale with a late hop addition (2.5 ml oil/hl) and one half dry hopped in addition (1.5 ml oil/hl). The analytical differences in the hop samples could be followed through in the beer. Thus the beers brewed with Hallertau hops had significantly higher polyphenol contents, the American hops on the other hand more linalool. The higher ester content of the HCA is reflected in the beers.
No great differences were found in the tastings made by various panels.

Descriptors: hops, late hopping, dry hopping, hop growing region, cascade, comet, provenience

1 Introduction

In the 19th century and in the first half of the 20th, the location and region of where hops were grown played a much greater role in their evaluation than today. It is documented, for example, that the *Hallertauer Mittelfrüh* variety, introduced into the Spalt growing region in the 19th century, underwent a sensory modification and came closer in aroma to the Spalt variety [1].

Still after World War II great significance was given to a seal district in the Hallertau. Core seal districts like Au, Mainburg and Wolnzach were valued higher than outlying districts and there were many breweries who wanted hops only from a specific seal district.

Whereas the growing area played an important role about 100 years ago [2], in recent times there has been but little discourse on this topic that is backed by analytical data. Even the book "Hops – Their Cultivation, Composition and Usage", published in 2014, said very little about this, which is a pointer to its neglect in current literature [3].

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There is just one work from 2002, which for the first time proved systematic differences between the growing regions of Yakima (USA) and the Hallertau (Germany) based on the analysis of low-molecular polyphenols in the *Perle* and *Nugget* varieties using high-performance liquid chromatography (HPLC). In three crops (1996-1998) the Hallertau hops showed a higher polyphenol content than those grown in America. Differences were found both in substance groups and in single components [4].

In the meantime, there is an increasing trend towards questioning the influence of the growing region. Thus craft brewers are concentrating more on the natural fluctuations in aroma, for example, due to different stages of maturity of the hops [5]. However, aroma fluctuations from region to region are also being monitored [6].

In parallel, hop varieties with special aromas – also known as flavour hops – for dry hopping are catching the attention of small breweries (craft brewers) in particular. Here, the American breed *Cascade* is a kind of "lead variety". It was approved as an aroma variety in 1972 and, after almost completely disappearing, is now grown in considerable quantities in the USA (2,157 hectares in 2013). Since 2010, the *Cascade* variety has also been grown in the Hallertau. It currently covers an area of about 10 ha with a quickly rising tendency.

It was therefore interesting to see whether and if so, which differences there are between hops grown in Yakima and those grown in the Hallertau and the beers made of them. Are these differences significant enough to influence analytical and sensory properties of the beers and thus complicate substitution? The first results from the 2012 crop have already been presented in a poster [7].

The situation is similar with the *Comet* variety which is grown in small quantities in both the USA and the Hallertau. *Comet*, like *Cascade*, engendered from the USDA breeding program at the Oregon State University (OSU), Corvallis, and was registered as a high-alpha variety in 1962. The *Comet* variety attained its peak in 1980 with a growing area of 260 ha, but was then quickly overtaken by other varieties with an even higher alpha content and is now hardly grown at all commercially in the USA (9 ha in 2013), beside the fact of an very interesting aroma.

The present paper compares the *Cascade* variety as grown in the USA (Yakima) = US-CA, and in Germany (Hallertau) = HCA, taking the crops of 2012 and 2013. Only the 2013 crop was available from the two growing areas for analysis of the *Comet* variety (US-CM and HCM respectively).

2 Test Program

The vegetation periods of 2012 and 2013 ran counter to each other in Yakima and the Hallertau. Whereas in 2012 there were favorable climatic conditions in the Hallertau and in 2013 rather unfavorable conditions, it was the reverse case in Yakima.

2.1 Hop Samples and Analysis

For the program *Cascade* hop samples were chosen in crop 2012 and 2013, *Comet* in 2013 only. The hops were supplied by John I. Haas and HVG.

The analysis of the samples was focused on the following:
- Bitter substances according to EBC 7.7, alpha and beta acids
- Aging index = Hop Storage Index (HSI according to ASBC)
- Total oil according to EBC 7.10
- Total polyphenols according to an "AHA" method
- HPLC polyphenols
- Aroma components analyzed using GC-FID
- Aroma substances with sulfur content

The Hop Storage Index was determined in each case to ensure that the aging of the US and Hallertau samples was comparable.

Analysis of the total polyphenols was done using a non-specific method of the AHA (Hop Analysis Working Group), which is based on the hot water extraction of the hop sample. The solution is filtered and then a spectrophotometric measurement is done similar to the EBC 9.11 method.

Low-molecular polyphenols can be separated by means of HPLC and quantified to a great extent [3, 4]. Hop samples are extracted in acetone/water and residual resins are removed with hexane. The polyamide adsorbed polyphenols are washed out with methanol and the solution is put in a gradient HPLC system with diode array detector.

Column: Phenomenex Luna 5µ (18/2) 100A, 250 x 4,6 nm

Solvent: Methanol and alkaline methanol

Gradient:	A	Water / formic acid	95/5
	B	Methanol/acetonitrile/formic acid	95/5/5

Single components are summarized in the following groups:
- Hydroxycinnamic acids
- Monomeric flavanols
- Proanthocyanidins
- Quercetin flavonoids
- Kaempferol flavonoids
- Other flavonoids
- Sum of all the analyzed HPLC substances

The aroma components for the Cascade crop of 2012 resulted from one laboratory (Nateco$_2$, Wolnzach). Whereas the process for obtaining the oil and separating the single components according to EBC involves 3 hours of distillation at approx. 100°C followed by GC-FID analysis, here a gentler method of quick extraction lasting 15 minutes was applied acc. MEBAK III 1.4, using a Büchi K-355 still. The hop oil is separated into a hydrocarbon fraction and an oxygen fraction and these two fractions are analyzed separately by GC-FID. The four samples from the 2013 crop were analyzed as well in another laboratory [KU Leuven, Technology Campus Ghent, Laboratory of Enzyme, Fermentation and Brewing Technology (EFBT)], via headspace solid-phase microextraction and gas chromatography-mass spectrometry which besides providing a countercheck also included additional terpene alcohols and their esters described in [8].

Aroma components with sulfur content play a major role in flavour hops. In particular, with 4-sulfanyl-4-methylpentan-2-one (4MMP or 4S4M2Pone), Cascade has a highly active sensory substance. We will not go into detail about the relevant literature here. Important references are to be found in [9, 10]. The analyses were done in an institute of the Université Catholique de Louvain. The methods applied are described in essence in [11, 12]. Polyfunctional thiols were extracted with pHMB and analyzed in a GC with pulsed-flame photometric detector (PFPD).

2.2 Brew Program

The hop samples were brewed in the 2 hl research brewery in St. Johann, Germany. The beers brewed with hops of the 2012 crop received pellets of the Herkules variety at the beginning of boiling to give the desired bitterness. The 2013 crop samples were brewed with single varieties and dosed at the beginning of boiling and at the end of boiling and dry hopped in addition. In the following means regardless a first addition in every brew:
- "late" = hop addition at the end of boil / whirlpool
- "dry" = late + an additional dry hopping

The main characteristics of the trial brews are as follows:
- Bottom fermenting lager
- 100 % Pilsner malt
- Original extract: approx. 12 %
- Bitterness: 25 IBU

Table 1 Hopping of the beers with samples from the 2012 crop (1st addition Herkules); values in g/hl

	US-CA		HCA	
	late	dry	late	dry
Begin of boil (Herkules)	129	129	108	108
End of boil (2.5 ml oil/hl)	200	200	161	161
Dry hopping (1.5ml oil/hl)	–	120	–	97

Table 2 Hopping of the beers with samples from the 2013 crop (single varieties); values in g/hl

	US-CA		HCA		US-CA		HCA	
	late	dry	late	dry	late	dry	late	dry
Begin of boil	103	103	135	135	73	73	84	84
End of boil (2.5 ml oil/hl)	147	147	217	217	135	135	179	179
Dry hopping (1.5ml oil/hl)	–	88	–	130	–	81	–	107

Table 3 Content of α-acids, polyphenols, xanthohumol and hop oil as well as the following ratios β:α, cohumulone level, polyphenols:α, xanthohumol:α, hop oil:α and Hop Storage Index; US-CA and HCA 2012 and 2013 crops; US-CM and HCM 2013 crop

		Crop 2012		Crop 2013			
		US-CA	HCA	US-CA	HCA	US-CM	HCM
α-acids	% w/w	6.2	6.7	7.3	5.6	8.8	7.7
β : α	–	0.95	0.94	0.97	1.13	0.39	0.62
Cohumulone Ratio	% rel	32	34	32	33	36	40
Hop Storage Index	–	0.33	0.27	0.27	0.26	0.32	0.28
Total Polyphenols	% w/w	3.5	4.9	3.5	4.6	2.6	4.5
Xanthohumol	% w/w	0.30	0.45	0.32	0.40	0.86	0.80
Hop Oil	ml/100g	1.25	1.55	1.70	1.15	1.85	1.40

The 4 beers of the 2012 crop and the 8 beers of the 2013 crop were hopped with the doses shown in tables 1 and 2.

The addition of hops at the beginning of boiling was to obtain the desired bitterness of 25 IBU taking into account the yield in the last addition of about 5 %. The late addition with the hop equivalent of 2.5 ml hop oil/hl was done at the end of boiling and in the whirlpool at a ratio of 1:1.

After the primary fermentation half of each of the beers was dry hopped with the trial hops at a volume equivalent to 1.5 ml hop oil/hl. The residence time was 1 week at 14°C and 2 weeks at 0°C, which permits an intensive extraction of substances and also does not exclude reactions with yeast enzymes. The beers that were purely late hopped and the ones that were additionally dry hopped were bottled separately after filtration. A control beer for the beers in table 1 was bittered with Herkules pellets only at the beginning of boiling.

2.3 Beer Analyses

The analyses of the beers essentially covered the following characteristics.

- General beer analyses (OG, ADF, pH) primarily for checking the reproducibility of the brews
- Bitterness units according to EBC 9.8 (IBU)
- HPLC bitterness components (α-acids and iso-α-acids) according to EBC 9.47
- Total polyphenols according to EBC 9.11
- Linalool using GC-FID (Nateco$_2$ laboratory)
- Aroma substances using SPME-GC-MS (Ghent laboratory)

2.4 Sensory Analysis of the Beers

The beers were tasted by the panel of the St. Johann research brewery (7–12 trained tasters) and by a consumer panel of 38. The following criteria were judged:

- Intensity and quality of the hop aroma
- Intensity and quality of the bitterness
- Body

In addition, personal preferences were asked. The panel in St. Johann also had the task of assessing specified aroma impressions (fruity, floral, citrusy, herbal and hoppy).

The dry-hopped *Cascade* beers from the 2013 crop were presented at the 7th Workshop held by the "Beer Lateral Thinkers" (Bier-Quer-Denker) on 02. 04. 2014 in Nuremberg. The 118 participants evaluated the beers according to the following criteria:

- Intensity of hop aroma from 1 to 10
- Quality of hop aroma from 1 to 10
- Quality of the bitterness (preference between Beer 1 and Beer 2 only)
- Generally preferred beer

As the quality of hop aroma and bitterness cannot be measured objectively the evaluation of these criteria follows a hedonistic approach.

3 Results

3.1 Hop Analyses

3.1.1 General Hop Analyses

In the 2012 crop, the α-acids and oil of the Hallertau *Cascade* were superior to the US Cascade; the reverse was true in the 2013 crop. The polyphenol content was not noticeably influenced by the year. The HCA was ahead in both crops with 44 rel. % and 31 rel. % respectively. The ratio hop oil:α showed no influence either by year or location (Table 3).

Table 4 Low-molecular polyphenols analyzed by HPLC in mg/100 g; *Cascade* 2012 and 2013 crops; *Comet* 2013 crop

	Crop 2012		Crop 2013			
	US-CA	HCA	US-CA	HCA	US-CM	HCM
Hydroxy cinamic acids	109	188	104	200	58	181
Flavanols	94	110	104	187	87	167
Proanthocyanidines	137	172	132	250	75	190
Quercetin flavonoides	207	340	215	394	178	375
Kaempferol flavonoides	171	258	105	219	53	118
Other flavonoides	13	24	17	29	28	51
Sum of HPLC Polyphenols	840	1256	826	1472	625	1282

The Hallertau *Comet* had less α-acids and hop oil than the American. In contrast, the HCM has a 73 % greater polyphenol content than the US-CM, which leads to the conclusion that the observations made in [4] about the *Perle* and *Nugget* varieties are also relevant for *Cascade* and *Comet*.

The prenylflavonoid xanthohumol differs from most other polyphenols. Its biogenesis is linked more to the development of the α-acids. It does not dissolve well in water and it is contained in the lupulin glands along with the hop resins and oil, unlike the majority of polyphenols which are located in the bracts [3]. In the case studied, the α-richer *Comet* had twice as much xanthohumol than the *Cascade*. The ratio xanthohumol:α was greater in the Hallertau samples. However, it was reported also in [4] about higher xanthohumol contents in Hallertau *Perle* and *Nugget* compared with the American hops.

Table 5 Relative additional quantity of polyphenols (total and HPLC) in Hallertau hops compared with US hops

		Total Polyphenoles	HPLC Polyphenoles
Cascade	Crop 2012	+44	+50
Cascade	Crop 2013	+31	+78
Comet	Crop 2013	+73	+105

The two *Cascade* hops were not exact the same in Hop Storage Index in the 2012 crop with 0.33 (US) and 0.27 (HAL), but were in the 2013 crop (0.27 and 0.26 respectively).

In the case of *Comet*, the difference between US-CM (0.32) and HCM (0.28) was similar bearing in mind that it is almost impossible to get hop samples from different regions with an identical HSI.

3.1.2 Low-molecular Polyphenols

Table 4 lists the low-molecular polyphenols analyzed by HPLC as sums in substance groups. The valuable role played by low-molecular polyphenols – as a flavour carrier, in the body or as polyphenolic bitter substances in the harmonious rounding-off of the beer bitterness – is presented more detailed in [3].

Table 5 gives a summary of the additional quantities in the Hallertau hops compared with the US hops in relative %.

3.1.3 Aroma Components

Hop aroma components are analyzed using GC-FID in conjunction with MS. The hydrocarbon fraction (HCF) and the oxygen fraction (OF) are measured separately. Table 6 shows some of the main aroma substances of all six samples.

With *Cascade* there is a tendency similar to the α-acids. With the 2012 crop the Hallertau hop is slightly ahead of the US sample, which is due to the favorable weather conditions. With the 2013 crop

Table 6 Selected aroma components in mg/100 g; *Cascade* 2012 and 2013 crops; *Comet* 2013 crop

	Crop 2012		Crop 2013			
	US-CA	HCA	US-CM	HCM	US-CA	HCA
Myrcene	640	720	652	609	863	595
α-Ocimene	2	4	18	13	3	3
β-Caryophyllene	50	48	82	148	60	39
β-Farnesene	51	66	9	11	61	58
α-Humulene	115	134	11	29	126	110
α- + β-Selinene	16	14	89	128	20	17
γ- + δ-Cadinene	8	8	3	5	9	7
Selina (4,7) + (3,7)-diene	0	0	14	23	0	0
Isobutyl isobutyrate	2	4	5	4	3	5
Isoamyl propanoate	4	4	17	4	5	2
3-Methylbutyl 2-methylpropanoate	3	3	4	2	4	3
2-methylbutyl 2-methylpropanoate	9	17	17	12	13	17
Linalool	6	5	10	6	8	3
2-Undecanon	2	2	2	3	2	1
Methyl 4-decenoate	4	4	8	6	4	3
Methyl 4,8-decadienoate	2	5	4	2	3	2
Geranyl acetate	14	15	11	18	11	12
Methyl dodecadienoate	2	1	10	13	8	8
Geraniol	5	7	5	7	14	7
Epoxides (Car.I,Hum I,II,III)	21	8	9	6	7	4
Selina-11-en-4-ol	3	1	7	8	1	1
Hydrocarbon fraction (HCF)	903	1014	905	990	1164	846
Oxygen fraction (OF)	106	105	163	138	105	82
Ester ratio	17	27	26	15	24	33
Epoxides : Caryoph. + Humulene	13	4	10	3	4	3
HCF : OF	8.5	9.7	5.6	7.2	11.1	10.3
Humulene : β-Carophyllene	2.3	2.8	0.13	0.2	2.1	2.8

Table 7 Aroma substance with sulfur content in the *Cascade* samples of the 2013 crop (values in µg/kg)

	2013	
	US-CA	HCA
unknown 1	38	89
4-Sulfanyl-4-methylpentan-2-one	32	87
unknown 5	16	16
3-Sulfanyl-3-mythylbutan-1-ol	21	29

Table 8 Terpene alcohols and their esters in US and Hallertau *Cascade*; 2012 and 2013 crops; parts of the data from 2 laboratories; values in mg/100g; na = not analyzed

	Crop 2012				Crop 2013			
	US-CA		HCA		US-CA		HCA	
	EFBT	NA	EFBT	NA	EFBT	NA	EFBT	NA
Linalool	7.6	6	6.6	5	11.2	8	5.0	3
Geraniol	2.5	5	7.6	7	14.2	14	7.4	7
Nerol	< 0.1	–	0.2	–	< 0.1	–	< 0.1	–
Geranyl acetate	8.0	14	10	15	9.2	11	9.1	12
α-Terpineol	0.2	–	0.2	–	0.2	–	0.1	–
β-Citronellol	0.3	–	0.5	–	0.6	–	0.3	–
Geranyl propanoate	na	–	na	–	1	–	0.5	–
Geranyl isobutyrate	na	–	na	–	2.3	–	1.8	–

the US-CA comes out on top. This observation is derived from most of the single components and clearly from the sum of the hydrocarbon fraction and the oxygen fraction.

The following provenience-specific observations are interesting:

- Linalool is present in greater quantities in the US-CA hops of both crops.
- Geraniol is twice as high in the US-CA hops of the 2013 crop.
- There are clearly higher quantities of the esters isobutyl isobutyrate and 2-methylbutyl 2-methylpropanoate in the Hallertau *Cascade* of both crops than in the US samples.

In total with the exception of linalool, the HCM has slightly higher single values than the US-CM of the 2013 crop. However, a clear influence of the growing region cannot be derived from this.

Since the results of sulfuric compounds in the *Cascade* samples of the 2012 crop were not very conclusive, table 7 lists only the two *Cascade* hops of the 2013 crop. This comparison shows that the Hallertau pellet has higher values than the US pellet. This is the first indication that the variety-typical substances with sulfur content in the Hallertau *Cascade* are at least on a par with those of the US-CA.

If you gather all the analytical data together and try to eliminate the yearly fluctuations, the total polyphenols, in particular the low-molecular HPLC polyphenols, can be taken to be safe indicators of the influence of the growing regions. In all cases the Hallertau hops have considerably higher values than the American hops.

The linalool values are higher in the US hops of both crops and varieties. The Hallertau *Cascade* has significantly higher values for two esters in both crops. Table 8 gives an overview of the terpene alcohol and ester data from $Nateco_2$ (NA) and KU Leuven, Technology Campus Ghent, Laboratory of Enzyme, Fermentation and Brewing Technology (EFBT)

Considering the difficulties of the analysis, it can be established that a comparison of the data from the two laboratories is satisfactory. What is interesting, however, is that, at least with *Cascade*, the substances nerol, α-terpineol, β-citronellol and geranyl propanoate do not play a significant role compared with linalool, geraniol and geranyl acetate. At most there is slightly more geranyl isobutyrate which might contribute to a certain extent to the total geraniol in beer. However, the quantities here do not correspond quite to those given in [13].

3.1.4 Sensory Characterization of Cascade

Hops have long been evaluated in a sensory manner, i. e. through smelling. There are numerous suggestions for describing this, but they cannot be compared with each other. The authors of the book about hops which has just been published have therefore refrained from using sensory descriptions in presenting the hop varieties [3].

Almost at the same time a multivolume work was published describing all the hop varieties that are currently being grown commercially around the world [14]. Volume 3 includes descriptions of the *Cascade* variety as grown in various regions. The sensory assessment is nevertheless based on 11 olfactory characteristics

Table 9 Seven sensory characteristics of *Cascade,* grown in USA and in the Hallertau; scores of 0 to 9

	USA	HAL
Caramel	7	0
Herbal	6	3
Red Berrys	8	6
Sweet Fruits	5	5
Floral	2	3
Green Fruits	1	6
Citrussy	6	8

Table 10 Analyses of the 5 test beers (incl. comparison) with specification of the relevant difference between late and additionally dry hopped beers

		Control	US-CA		HCA		Difference
		*	late	dry	late	dry	dry/late
Original Gravity	% w/w	11.8	12.0	12.4	12.0	12.3	+3%
Bitterness	IBU	23	27	31	24	27	+7%
Iso-α-acids	mg/l	23.8	18.8	19.5	19.6	20.5	+4%
α-acids	mg/l	0.6	3.5	5.0	3.7	5.2	+42%
Total Polyphenols	mg/l	141	182	212	205	240	+17%
Hop Polyphenols	mg/l	–	41	71	64	99	+62%

* hop addition only begin of boil

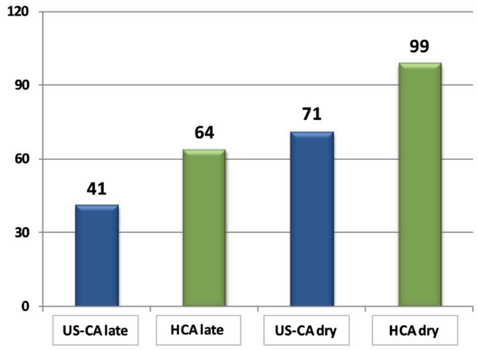


Fig. 1 Additional quantity of polyphenols in the 4 test beers; comparison of US-CA and HCA; late and additional dry hopping; values in mg/l

with scores from 0 to 9, from indistinct to very distinct. The data in the spider diagram has been converted into table form for the 7 main characteristics of US-CA and HCA (Table 9).

The differences in the scores for the characteristics of creamy caramel, spicy and green fruits were enormous, whereas characteristics of red berries, sweet fruits and citrus were better matched. However, the analyses of the aroma substances from both crops would give good reason to expect a generally better match of the sensory evaluations. This underlines the problem of sensory assessments which only represent a snapshot with all the associated imponderables like using a single sample, selection of test persons, how they feel on the day and subjective impressions.

3.2 Beer Analyses

Since beers with hops from the 2012 crop were bittered with pellets of the *Herkules* variety in the 1st addition at the beginning of boiling, and those with hops from the 2013 crop were single hop brews, the two crops can only be discussed separately.

3.2.1 Beers with Cascade from the 2012 Crop

Table 10 lists the major general beer analyses of the control beer and the 4 trials with *Cascade*.

The following differences are noticeable:

- The polyphenol content of the HCA beers is significantly greater than that of the US-CA beers.
- The observations made earlier about dry hopping are confirmed [15]: Iso-alpha-acids remain constant, whereas alpha acids and consequently the bitterness units increase. Also the polyphenols rise by dry hopping with pellets.

If you subtract the 141 mg/l of polyphenols of the control beer from the polyphenol content of the 4 test beers, you get the additional quantity of hop polyphenols from the late addition and the dry hopping. This is shown in figure 1.

Table 11 Polyphenol dosages in mg/l through late and dry hopping as well as the transfer rates of the polyphenols (additional quantity in the beer : dosage) in rel. %

		US-CA	HCA
Dosage of Polyphenols	late mg/l	68	79
	dry mg/l	41	48
Transfer Rate of Polyphenole	late % rel.	60	81
	dry % rel.	73	73

Table 12 Selected aroma components in the 4 test beers of the 2012 crop and the control beer in µg/l

	Control	US-CA		HCA	
	*	late	dry	late	dry
Myrcene	nd	1.1	1.7	1.0	2.0
Linalool	4	101	145	82	123
3-Methylbutyl 2-methylpropanoate	nd	7	19	8	22
2-Methylbutyl 2-methylpropanoate	nd	41	106	79	198
Isobutyl isobutyrate	nd	5	12	13	32
Isoamyl propanoate	nd	15	35	17	36
Sum of Esters	–	68	172	117	288

* hop addition only begin of boil

The dosed quantities of polyphenols can be calculated from the polyphenol content of the hops (Table 3) and the hopping (Table 1). These values in relation to the additional quantities in the beers (Table 10) lead to the transfer rates of the polyphenols in % relative (Table 11).

The calculated transfer rates are somewhat over the figures published in [3] and [15] but within the limits of error analysis.

Table 12 shows the values of a number of hop aroma components in the four beers. In the control beer only a small amount of linalool can be detected. There are noticeably higher values of isobutyl isobutyrate and 2-methylbutyl 2-methylpropanoate in the beers hopped with HCA, which is due to the corresponding differences in the hops. The higher linalool values in the US-CA beers come

Table 13 Dosages of some aroma substances in µg/l and their recovery percentage (relative %) through late and dry hopping

		US-CA		HCA	
		Dosage µg/l	Transfer rate % rel.	Dosage µg/l	Transfer rate % rel.
Linalool*	late	136	71	93	83
	dry	82	54	56	74
Isobutyl isobutyrate	late	40	18	64	20
	dry	24	50	39	49
3-Methylbutyl 2-methylpropanoate	late	60	12	48	17
	dry	40	30	29	55
2-Methylbutyl 2-methylpropanoate	late	180	23	274	29
	dry	108	60	165	72
Isoamyl propanoate	late	80	19	64	27
	dry	48	42	39	49

* Dosage is calculated with mean values of both labs; US-CA = 6.8 mg/100g; HCA = 5.8 mg/100g

Table 14 General beer analyses including the polyphenol data and the polyphenols transferred by the hops

		US-CA		HCA		US-CM		HCM	
	Unit	late	dry	late	dry	late	dry	late	dry
Original Gravity	% w/w	12.3	12.3	11.9	11.9	12.0	11.9	11.7	11.7
App. Degree of Fermentation	% w/v	82.3	84.8	82.5	85.7	81.7	84.7	82.8	84.0
pH		4.64	4.62	4.54	4.53	4.46	4.54	4.46	4.46
Bitter Units	IBU	27	28	23	27	22	27	22	24
Iso-α-acids	mg/l	21.4	21.2	20.5	21.2	17.4	17.0	18.7	17.6
α-acids	mg/l	4.7	5.1	3.5	5.9	3.5	6.5	2.9	4.8
Total Polyphenols	mg/l	197	209	232	257	179	185	203	221
Hop Polyphenols	mg/l	77	89	112	137	59	65	83	101

from the slightly higher value in the US *Cascade*, in particular from its higher dosage according to the oil content.

Similar to the polyphenols, the transfer rates of aroma substances can also be calculated. Only for linalool there was a blank value of 4 µg/l in the control beer, which has to be subtracted from the test brews values. From the additionally dry hopped beers you must subtract the value of the beers that were only late hopped. The dosages are calculated from the additions (Table 2) and the contents in the hops (Table 7). The transfer rates in relative % now can be calculated from the resulting values and the dosages (Table 13).

The linalool transfer rates are somewhat higher than usual for late hopping [3], but still within the high ranges of fluctuation, those for dry hopping are slightly below the data published to date [15]. The 4 esters are behaving practically uniform. Late hopping yields transfer rates of 12 % to 29 % (average 21 %); dry hopping yields transfer rates of 30 % to 72 % (average 51 %).

3.2.2 Beers with Cascade and Comet from the 2013 Crop

The standard beer analyses are given in table 14. An approximate calculation can be made as follows to determine the transferred by the hops. The control beer had 141 mg/l. The first addition of *Herkules* pellets transferred about 38 mg/l. At a standard yield of 55 % in the first addition [3] the result is 21 mg/l. The unhopped beer thus receives 120 mg/l of polyphenols. Table 14 shows also the hop polyphenols transferred in each beer.

The original extracts vary between 11.7 % and 12.3 %, the pH from 4.46 to 4.64, which indicates a good reproducibility of the brews. The degree of fermentation is worth noticing. It is striking that, with practically the same substrate, the dry hopped beers have higher apparent degrees of fermentation – on average 84.8 ± 1.1 % compared to 82.3 ± 0.7 % in beers which are only late hopped. It is conceivable that dry hopping introduces additional fermentable sugars which is not yet confirmed in literature but backed up by several own experiments.

The following statements confirm earlier observations [15]:

- The bitterness units of the additionally dry hopped beers are on average 3 IBU higher, indicating that bitter components are solubilized.
- Since no isomerization of α-acids can take place at beer storage temperatures, there is no difference in the values of the two series (late: av. 19.5 mg/l, dry: av. 19.3 mg/l).
- There is an explanation for the lower iso-α-acid values in the *Comet* beers. The lowest pH value during fermentation was 0.05 to 0.1 lower in the *Comet* beers and might therefore have favored secretion processes.
- α-acids dissolve in low amounts even with dry hopping (late: av. 3.7 mg/l, dry: av. 5.6 mg/l).
- As was expected from the hop data, all beers brewed with Hallertau hops stand out due to their higher polyphenol content. This becomes even clearer when considering the polyphenols introduced by the hops alone. In the case of the US hops the average amount is 73 mg/l and with the

Table 15 Hop aroma substances in the 8 beers

	US-CA		HCA		US-CM		HCM	
	late	dry	late	dry	late	dry	late	dry
Linalool	71	130	45	81	109	175	60	108
Geraniol	51	256	53	238	22	165	58	241
Nerol + Citronellol	18	46	15	29	10	28	18	26
Geranyl acetate	4	4	3	4	3	5	3	5
α-Terpineol	9	15	9	14	12	18	12	16
Myrcene	6	11	6	11	5	10	6	10
2-Methylbutyl propanoate	13	20	7	10	19	28	6	10
3-Methylbutyl 2-methylpropanoate	6	11	6	13	5	12	4	9
2-Methylbutyl 2-methylpropanoate	9	15	15	30	9	19	9	17
Non-sesquiterpenoid fraction	279	620	249	541	275	550	289	559
α-Selinen	1	2	1	3	14	28	12	25
Selina-3,7 (11) – diene	nd	nd	nd	nd	4	8	3	6
α- + β- + γ-Eudesmol	3	6	3	5	18	27	17	33
Caryophyllene + 3 Humulenepoxide	8	13	4	11	5	12	6	15
Sesquiterpenoid Fraction	64	119	47	86	144	249	136	275

Table 16 Dosed quantities and transfer rate of linalool ate and dry hopping

	Dosage µg/l		Transfer rate % rel.	
	late	dry	late	dry
HCA	64	99	62	90
USCA	103	165	52	84
HCM	83	134	53	104
USCM	109	175	52	79
Average			55	90

Hallertau hops it is 108 mg/l, i.e. about 50 % more.

- Taking the polyphenol transfer rates, the yield for the late hopped beers is 80 %, which is identical to the series of the 2012 crop. Dry hopping on the other hand gives a transfer rate of only 40 % compared with 60 % in beers from the 2012 crop.

Of the 59 analyzed aroma substances of the hops the most interesting found in the beer are chosen (Table 15): Five terpene alcohols, three esters, myrcene, four sesquiterpenoid substances and two sums (non-sesquiterpenoids and sesquiterpenoids).

The following observations are noted.

- The linalool values follow the hops. The US samples have a higher content, which is reflected directly in the beers. The linalool values of the dry hopped beers increase by 73 %.
- Despite a higher value in the hops the geraniol values of the *Cascade* beers are the same. The overproportional increase in geraniol through dry hopping, which does not correspond to the hop values, was explained in [16]. The high amounts of geranyl acetate in *Cascade* and *Comet*, are not found directly in the beers. Dosed geranyl acetate is hydrolyzed and thus increases the geraniol values. Geranyl isobutyrate reacts analogously and contributes to the geraniol yield [13].
- Nerol and citronellol are identical in the beers. The increase of 115 % through dry hopping is overproportional compared with linalool.
- α-terpineol is also similar in the beers. Dry hopping produces an increase of 50 %

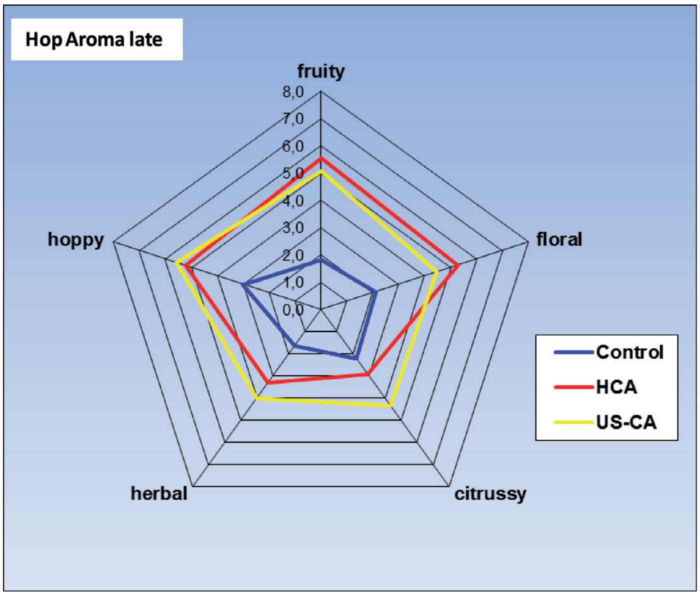
The following can be added regarding the behavior of the terpene alcohols nerol, citronellol and α-terpineol. The amounts of nerol, citronellol and α-terpineol in hops, as given in table 9, do not play a significant role compared with linalool, for example. In HCM, nerol content is only 3 % of the amount of linalool, citronellol an average of 6 % of the linalool and α-terpineol lies between 2 % and 3 %. In the beers, however, the ratio to linalool is on average 15 % for α-terpineol and 29 % for nerol + citronellol. The transfer rates are therefore significantly beyond 100 %.

The reasons for this might be transformation of the terpene alcohols as described in [17]. Thus, for example, citronellol can be formed from geraniol and α-terpineol from linalool. It is also possible that, with dry hopping, esters of the three alcohols hydrolyze like geranyl acetate. Neryl acetate is already detectable in some hops [18].

Further observations:

- Myrcene is an example of the low transfer rates of many monoterpenes and sesquiterpenes and does not play a sensory role.
- In the case of esters, the HCA beer reflects the higher content in the hops as shown in the 2012 crop.
- In the US-CA beer, the sum of the non-sesquiterpenoid fraction is 12 % (late) and 15 % (dry) respectively higher than in the HCA beers, which can be explained by the poor year due to unfavorable climatic conditions in the Hallertau.
- The sesquiterpenoids play a small role in quantity. The differences between the two *Cascade* beers are negligible. What is interesting here is the *Comet* which differs greatly from the *Cascade*. An influence of provenience cannot be clearly defined with the *Comet* beers.

Transfer rates for the linalool were calculated using the values of the Nateco$_2$ laboratory. Table 16 shows the transfer rates which match the data given in [3] and [15].



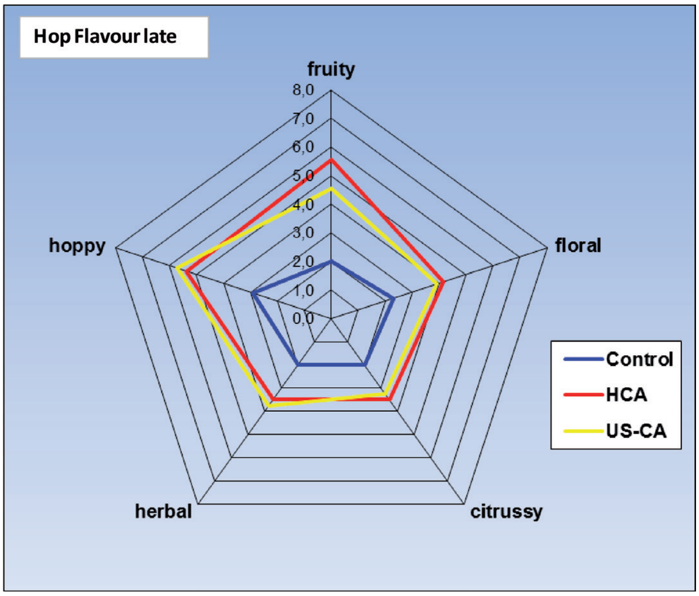


Fig. 2 Aroma and taste profiles for the late hopped beers including the control beer

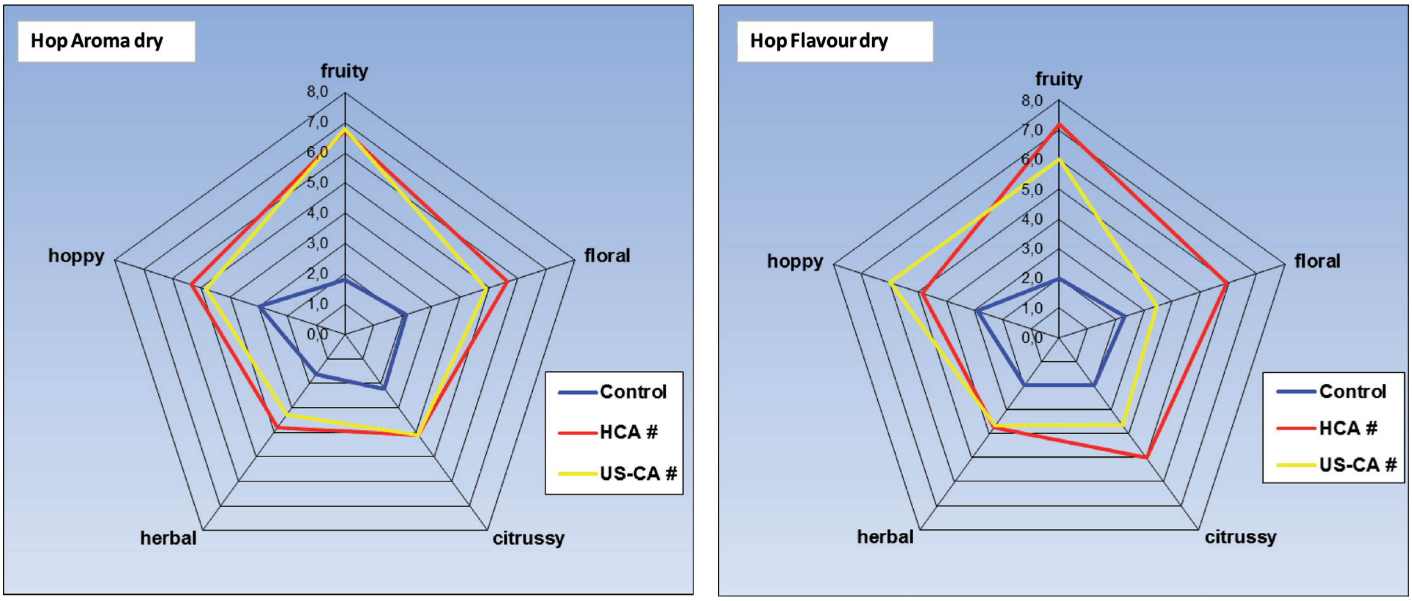


Fig. 3 **Aroma and taste profiles for the additionally dry hopped beers (#) including the control beer**

The differences in the hops from the two growing regions are reflected in the beers. It is essentially a matter of the polyphenols, linalool and two esters. Apart from that, there are no differences between the corresponding US and Hallertau beers.

3.3 Sensory Results

Here, too, the crops are treated separately, because the beers were not identically hopped in the 1st addition. Furthermore, the tastings were one year apart and the taste panels were not the same.

3.3.1 Beers with Cascade from the 2012 Crop

In the usual assessment of body and bitterness, the St. Johann taste panel could not establish any differences between the beers hopped with *Cascade* from the US and the Hallertau.

The sensory results (hop aroma descriptors) of the beers according to [19] are shown in figures 2 and 3. The aroma and taste impressions like hoppy, herbal, citrusy, floral and fruity had to be scored on a scale of 1 to 10. The more intensive impression given by the dry hopped HCA beer, particularly in flavour with the characteristics floral, fruity and citrusy, may be depending on the higher ester content of HCA.

A consumer panel of 38 could neither find any significant differences in intensity and quality of the hop aroma as well as bitterness nor in the body.

3.3.2 Beers with Cascade and Comet from the 2013 Crop

The results of the St. Johann taste panel are shown in the figures 4–7.

In assessing the intensity and quality of the hop aroma, Figure 4 shows that only the Hallertau *Comet* stands out from the late hopped beers, a fact which becomes less evident in the dry hopped beers. Whereas quality of the hop aroma of the late hopped HCA beers is slightly better than that of the US-CA beers, the tendency is reversed for the dry hopped beers. Unusual is the merely slight increase in the intensity of the hop aroma through dry hopping

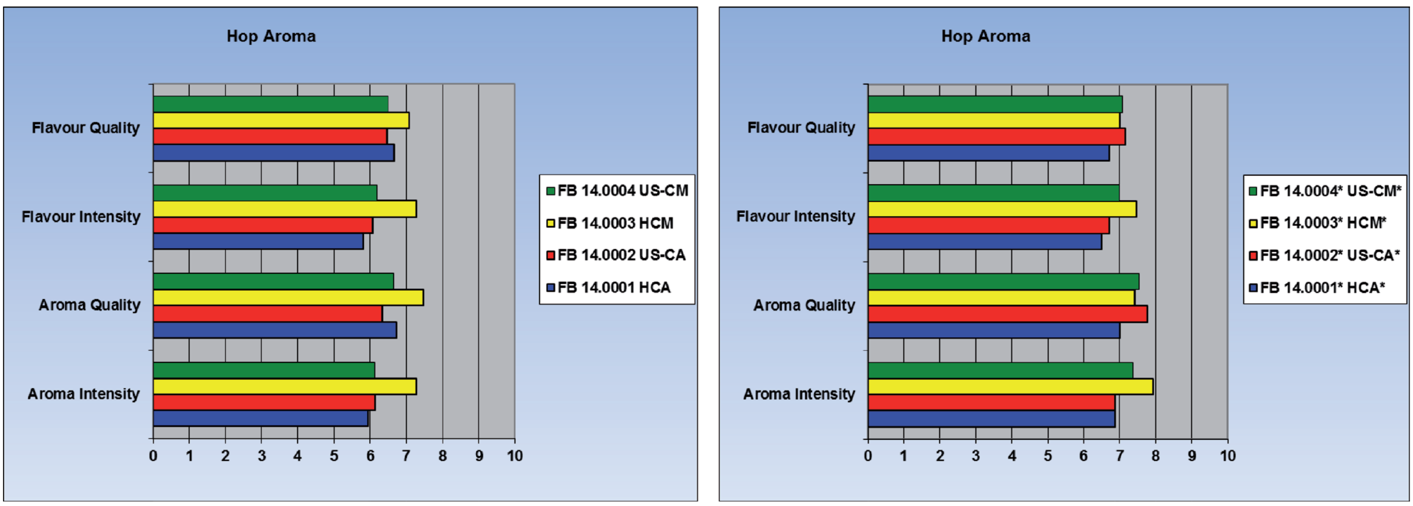


Fig. 4 **Intensity and quality of hop aroma and hop flavour (* = dry hopped)**

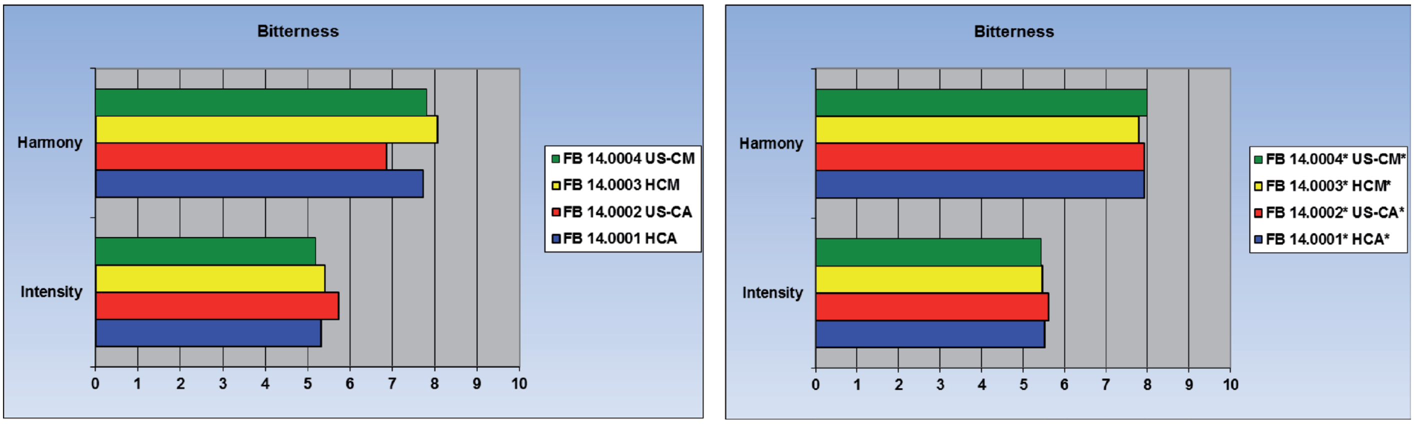


Fig. 5 Intensity and quality of the bitterness (* = dry hopped)

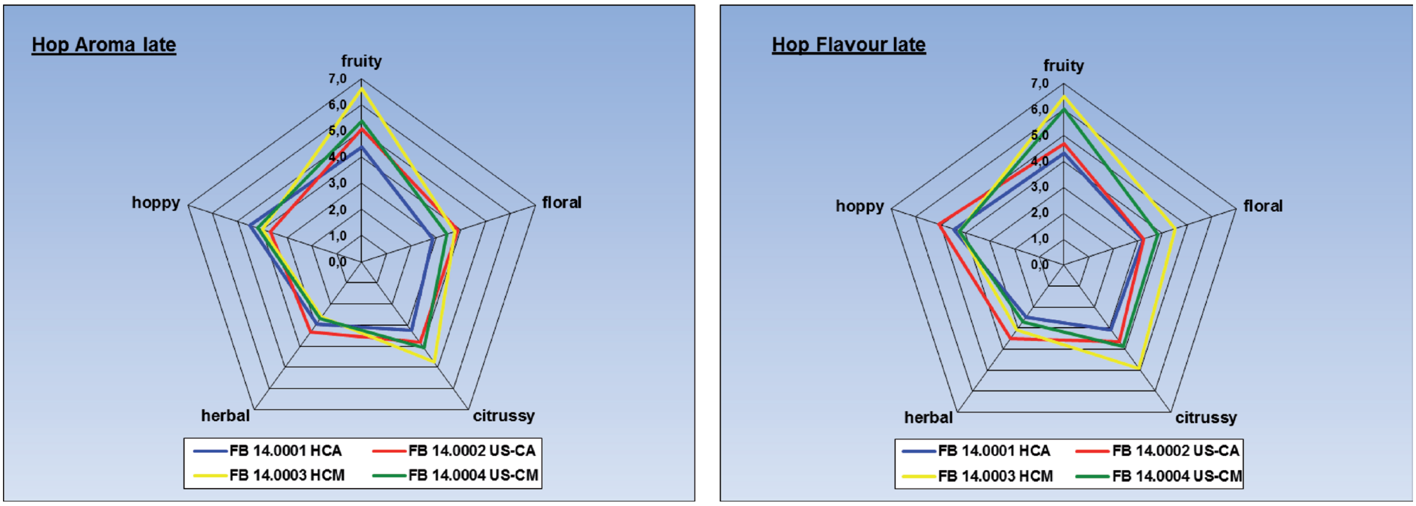


Fig. 6 Aroma and taste impressions of the 4 late hopped beers

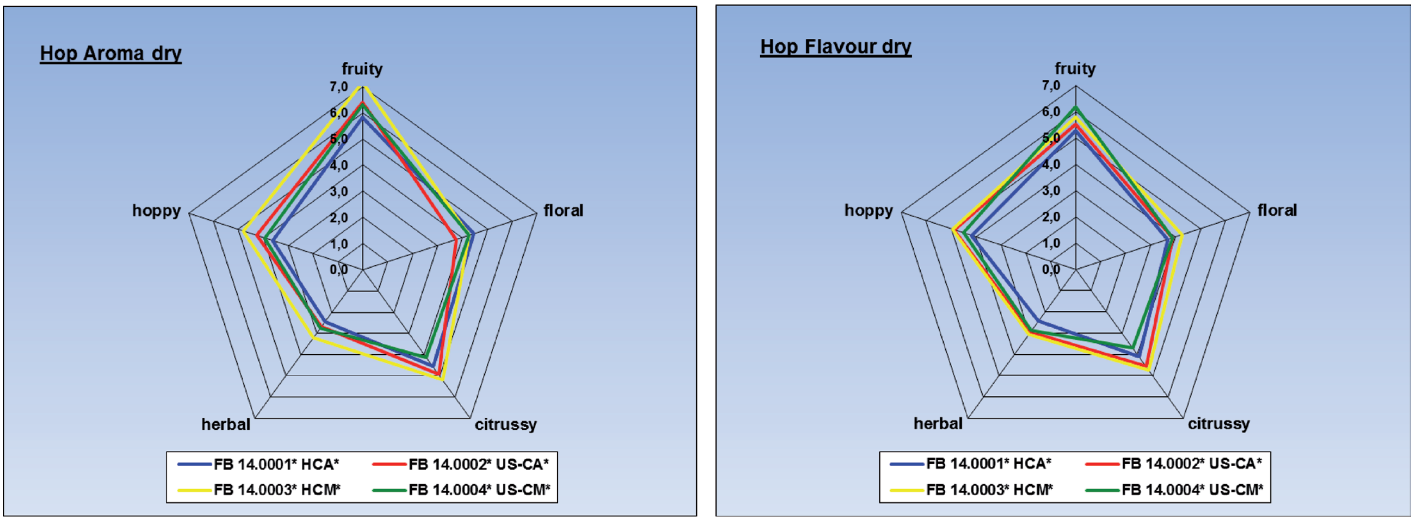


Fig. 7 Aroma and taste impressions of the 4 dry hopped beers

from an average of 6.4 points to 7.1 points.

The evaluation of the bitterness (Fig. 5) permits the following statements:

- The late hopped US-CA beer drops off slightly in the harmony of the bitterness.
- This tendency is lost in the dry hopped beers. The intensity and harmony of the bitterness in all four beers are practically the same.
- If you calculate the average values in the late and dry hopped beers, you get an intensity of 5.4 (late) compared with 5.5 (dry), and the harmony points of 7.6 (late) and 7.9 (dry) can be said to correlate. This once again proves that dry hopping with pellets does not have a negative but rather a slightly positive influence on the quality of the bitterness. This can be explained by additional hop polyphenols, which balance body and bitterness of the beers [3, 15, 20, 21].

Table 17 Average values for the interesting characteristics of fruity, floral and citrusy for aroma and flavour

		Aroma	Flavour	Average
US-CA	late	4.27	3.87	4.07
HCA	late	3.53	3.53	3.53
US-CA	dry	5.03	4.67	4.95
HCA	dry	4.97	4.37	4.67
US-CM	late	4.30	4.57	4.44
HCM	late	5.01	5.30	5.16
US-CM	dry	4.90	4.60	4.75
HCM	dry	5.53	4.93	5.23

Figure 6 illustrates the specific sensory impressions designated fruity, floral, citrusy, herbal and hoppy in the late hopped beers. There is hardly any difference in the herbal and hoppy characteristics of the 4 beers. In the other three characteristics the HCM beers are ahead, followed by the US-CM and the two *Cascade* beers.

The additionally dry hopped beers are all similar to each other (Fig. 7), although dry hopping with *Comet* had less effect than with *Cascade*.

Some average values for the interesting characteristics of fruity, floral and citrusy are given in table 17 for aroma and taste.

The following observations can be made:
- In both *Cascade* beers there is a clear increase in the average values of the characteristics going from the late hopped beers to the dry hopped beers.
- Dry hopping has less effect with *Comet*.
- The two *Cascade* beers show fewer differences between the growing regions than the *Comet* beers.

The tasters also had to give their preferences. There was no clear grading between the late and dry hopped beers:
- With the late hopped beers the Hallertau beers were all equally ahead of the US beers.
- With the dry hopped beers there were slight preferences for the US-CA ahead of HCM and HCA. US-CM came in last.

The tasting in pairs of the two growing regions resulted in two 1-star significant preferences for the beers that were only late hopped: HCA and HCM ranked each ahead of US-CA and US-CM. With the dry hopped beers US-CA was insignificantly preferred to HCA and HCM was insignificantly preferred to US-CM.

Table 18 Tasting results of the "Beer Lateral Thinkers" (intensity and quality scores 1–10)

	US-CA better than HCA		HCA better than US-CA		Total	
	n = 60		n = 58		n = 118	
	US-CA	HCA	US-CA	HCA	US-CA	HCA
Intensity of Hop Aroma	6.95	5.85	6.04	6.36	6.50	6.10
Quality of Hop Aroma	7.12	5.97	5.73	7.11	6.44	6.53
Ranking for Bitterness	1.15	1.85	1.88	1.12	1.51	1.49

In summarizing the results, there was no preference for one or other of the growing areas with *Cascade*. *Comet* develops an aroma in the beer which is similar to the characteristics in *Cascade*, but generally stronger. The HCM comes off better both in the late hopped (significant) and in the dry hopped (not significant) beer.

The consumer panel couldn't detect any significant differences.

The two dry hopped *Cascade* beers were presented for tasting at a "Beer Lateral Thinkers" event. Participants were asked to assess the intensity and quality of the hop aroma (scores of 1 to 10), and to give their preferences with regard to the bitterness and also their overall preferences.

60 participants preferred the US-CA beer, 58 the HCA beer. Table 18 gives a summary of the scores for intensity and quality of the hop aroma and the preference rank numbers for the bitterness separated according to preferences for one of the beers and as overall average values. The tasting results showed a high standard deviation.

Whereas the preferences of the single blocks in the evaluation are reflected, the overall average showed no significant differences, as expected. The intensity of the hop aroma in the US-CA beer is evaluated as marginally higher.

4 Summary

Already before 1900 hop varieties were transferred from one growing region to another, e.g. *Hallertau Mittelfrueh* to Tettnang and Spalt. In the 1980s, *Nugget* appeared in Europe and *Perle* in the USA. It was the great success enjoyed by *Cascade* as the "lead variety" for dry hopping in the American craft brewer scene that brought it to Germany. It is therefore of interest to discover whether there are differences between the *Cascade* hop grown in Yakima and that grown in the Hallertau.

To this end, a US *Cascade* (US-CA) was compared with a Hallertau *Cascade* (HCA) each from the 2012 and 2013 crops. The 2013 crop also provided a *Comet* from the USA (US-CM) and from the Hallertau (HCM). The hop samples were closely examined for aroma composition and polyphenols. Using the four *Cascade* samples and the two *Comet* samples, intensively hopped beers were brewed (late and dry hopping) and tested for analytical and sensory differences.

Systematic differences were found in both varieties with regard to polyphenols. The German *CA* and *CM* lie ahead of the American by 50 % and 78 % respectively with regard to total and low-molecular polyphenols including xanthohumol. Influences of crop years on the polyphenols are not given. All three US hops have higher linalool contents. There are clearly higher quantities of the esters isobutyl isobutyrate and 2-methylbutyl 2-methylpropanoate in the HCA of both crops. Variety-typical thiols (analyzed in the 2013 crop) were detected in both *Cascade* samples. The differences

in the other aroma components between the samples from both growing regions are to be considered low taking into account the usual fluctuations from lot to lot.

The six hops were brewed on the 2 hl scale with a late hop addition (2.5 ml oil/hl) and one half dry hopped in addition (1.5 ml oil/hl). The analytical differences in the hop samples could be followed through in the beer. Thus the beers brewed with Hallertau hops had significantly higher polyphenol contents, the American hops on the other hand more linalool. The higher ester content of the HCA is reflected in the beers. Although both *Comet* hops were also dosed by oil, the beers received a generally higher level of aroma substances. The established yields of polyphenols (late and dry, 50 % to 80 %), linalool (late about 50 %) and geraniol (over 100 % depending on the geranyl acetate content in the hops) confirm earlier results.

No great differences were found in the tastings made by various panels. Of the 2012 crop the HCA beer was considered fruitier, that of the 2013 crop was milder, which led to a preference for the late hopped beer. The same was the case with the HCM. The bitterness of the Hallertau beers was assessed to be slightly better, which is due to the higher polyphenol values.118 tasters compared the dry hopped *Cascade* beers of the 2013 crop. Despite a wide distribution of single assessments, the preference scores of 60 (US-CA) to 58 (HCA) produced a draw. There are definitely small sensory differences between the American and Hallertau beers, but these do not lead to any clear preferences.

Substitution of US by German hops of these varieties or vice versa is possible without changing substantial flavor characteristics in the beer.

5 Literature

1. Wagner, F.: Die bayerischen Hopfensorten, ihre Entstehung, Verbreitung und Eigenschaften; Verlagsbuchhandlung Eugen Ulmer, Stuttgart, 1905.
2. Strebel, E. V.: Handbuch des Hopfenbaues, Verlag von Eugen Ulmer, Stuttgart, 1887.
3. Biendl, M.; Engelhard, B.; Forster, A.; Gahr, A.; Lutz, A.; Mitter, W.; Schmidt, R. and Schönberger, C.: Hops – Their Cultivation, Composition and Usage, Fachverlag Hans Carl, 2014.
4. Forster, A.; Beck, B.; Schmidt, R.; Jansen, C. and Mellenthin, A.: Über die Zusammensetzung von niedermolekularen Polyphenolen in verschiedenen Hopfensorten und zwei Anbaugebieten; Monatsschrift für Brauwissenschaft **55** (2002), no. 5/6, pp. 98-108.
5. Bailey, B.; Schönberger, C.; Drexler, G.; Gahr, A.; Newman, R.; Pöschl, M. and Geiger, E.: The influence of hop harvest date on hop aroma in dry-hopped beers, MBAA Technical Quarterly, **46** (2009), doi:10.1094/TQ-46-2-0409-01.
6. Hieronymus, S.: For the love of hops; The practical guide to aroma, bitterness and the culture of hops; Brewers Publications, Boulder, Colorado, 2012, pp. 205-224.
7. Gahr, A. and Forster, A.: Comparison of Cascade Hops grown in the US and Germany, Poster no. 62, MBAA 2013 Annual Conference, October 2013, Austin, TX.
8. Van Opstaele, F.; De Causmacker, B.; Aerts, G. and De Cooman, L.: Characterisation of Novel Varietal Floral Hop Aromas by Headspace Solid Phase Microextraction and Gas Chromatography – Mass Spectrometry/ Olfactometry; J. Agric. Food Chem. **60** (2012), pp. 12270-12281.
9. Lermusieau, G. and Collin, S.: Volatile Sulfur Compounds in Hops and Residual Concentrations in Beer – A Review; J. of Am. Soc. Brew. Chem., **61** (2003), pp. 109-113.
10. Steinhaus, M. and Schieberle, P.: Transfer of the potent hop odorants linalool, geraniol and 4-methyl-4-sulfanyl-2-pentanone from hops into beer; 31st EBC Proc. 2007, Venice; paper 112.
11. Gros, J.; Nizet, S. and Collin, S.: Occurence of odorant polyfunctional thiols in the super alpha Tomahawk hop cultivar. Comparison with the thiol-rich Nelson Sauvin bitter variety, J. Agric. Food Chem., **59** (2011), pp. 8853-8865.
12. Vermeulen, C.; Lejeune, I.; Tran, T. and Collin, S.: Occurrence of polyfunctional thiols in fresh lager beers , J. Agric. Food Chem., **54** (2006), pp. 5061-5068.
13. Peacock, V. E.; Deinzer, M. L.; Likens, S. T.; Nickerson, G. B. and McGill, L. A.: Floral hop aroma in beer; J Agric. Food Chem., **29** (1981), pp. 1265-1269.
14. Joh. Barth & Sohn, The Hop Aroma Compendium, Vol. 1–3, Fachverlag Hans Carl, Nürnberg, 2011–2013.
15. Forster, A. and Gahr, A.: On the Fate of Certain Hop Substances during Dry Hopping, BrewingScience – Monatsschrift für Brauwissenschaft, **66** (2013), pp. 93-103.
16. Forster, A.; Gahr, A. and Opstaele, F. Van: On the Transfer Rate of Geraniol with Dry Hopping, BrewingScience – Monatsschrift für Brauwissenschaft, **67** (2014), pp. 60-62.
17. Takoi, K.: Biotransformation of monoterpene alcohols by lager yeast and their contribution to the citrus flavour of beer, paper 40 at ASBC Annual Meeting, Sanibel, Florida 2011.
18. Coelhan, M.: Verbesserung der Aromacharakterisierung der neuen Hueller Special Flavor-Hopfen; Internal Report, 15.11.2013.
19. "The spirit of beer – hops from Germany", CMA Hop Variety Portfolio, Centrale Marketing-Gesellschaft der deutschen Agrarwirtschaft mbH, Bonn, Germany, 2005.
20. Forster, A. and Gahr, A.: Hopping of Low Alcohol Beers; BrewingScience – Monatsschrift für Brauwissenschaft, **65** (2012), pp. 72-81.
21. Forster, A. und Gahr, A.: The Influence of Hopping on Beer Quality, BRAUWELT International, **29** (2011), pp. 19-23.

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