

# New International Calibration Standards ("ICS-2" Series) for HPLC Analysis of Iso- $\alpha$ -Acids and Tetrahydroiso- $\alpha$ -Acids

Submitted on behalf of the International Subcommittee  
for Isomerized Hop  $\alpha$ -Acids Standards  
by Richard Wilson, Chairman

The *International Subcommittee for Isomerized Hop  $\alpha$ -Acids Standards* (ISIHAS), announces the release of new calibration standards, ICS-2 and ICS-T2, for the HPLC analysis of iso- $\alpha$ -acids and tetrahydroiso- $\alpha$ -acids.

## Background

In April 2001, the ASBC, EBC, IoB (now IGB), and BCOJ approved the release of a set of HPLC standards for use in the quantitative determination of isomerized and reduced-isomerized  $\alpha$ -acids in hop products and in beer. These International Calibration Standards (ICS) were produced, analyzed and verified for the benefit of the brewing industry by the (ISIHAS) under the Chairmanship of Dr Richard Wilson (S.S. Steiner, Inc.).

Four standards were produced:

- DCHA-Iso, **ICS-I1** (Iso- $\alpha$ -acids standard);
- DCHA-Rho, **ICS-R1** (Rho-iso- $\alpha$ -acids standard);
- Tetra, **ICS-T1** (Tetrahydroiso- $\alpha$ -acids standard);
- DCHA-Hexa, **ICS-H1** (Hexahydroiso- $\alpha$ -acids standard).

The purity of each standard was determined using various HPLC procedures, elemental analysis and other methods. In each case, their total content of major isomers and homologs was declared and, before release, their stability was assessed and recommendations made regarding their storage and method of use. An isocratic version of EBC Method 7.8 (expected shortly to become EBC 7.9) was recommended as a convenient and generally applicable HPLC method for use in the analysis of unknown samples containing isomerized or reduced isomerized  $\alpha$ -acids.

Over a period of 2½ years, the subcommittee monitored the stocks of the four standards via careful HPLC analysis, finally concluding that the standards had maintained their declared compositions and had not undergone significant chemical change.

## Release of ICS-2 Standards for "Iso" and "Tetra"

The stocks of the DCHA-Iso (ICS-I1) and Tetra (ICS-T1) standards are now close to exhaustion. In anticipation of this, in 2003 the subcommittee initiated the preparation of replacement standards, which task is now completed. These standards were prepared and packaged by Dr John Paul Maye (John I Haas, Inc.) and his assistant, Susan Mulqueen, for which task the subcommittee records its considerable appreciation. Following their preparation, subcommittee members conducted extensive analysis in order to validate the standards and assign values to the content of their major components, this work including a collaborative HPLC study in which the prospective new standards were tested against the existing standards.

*These new standards, ICS-I2 and ICS-T2, now become the recommended standards and should be used for commercial transactions as well as for quality control purposes.*

## Use of the New Standards

As would be expected, the compositions of ICS-I2 and ICS-T2 are not identical to the standards they replace, but they can be used in exactly the same way. Dependent upon circumstances, users may find that the results for an unknown sample may differ slightly according to whether the old or the new standard has been used. In most cases, the differences observed when quantifying iso- $\alpha$ -acids using ICS-I2 instead of ICS-I1 will be found to be slight and usually within the normal range of experimental error. However, when using the new Tetra standard (ICS-T2) users may notice a small shift in the total "Tetra" value of an unknown sample previously analyzed using ICS-T1. The magnitude of this shift will vary according to the actual isomeric (*cis*- and *trans*-) and homological (co-, n- and ad-) composition of the unknown sample, but can be

expected to be a relative drop of around 1–2% in most cases (i.e. the new standard gives a lower result). However, the subcommittee is agreed that because ICS-T2 has a substantially higher *cis:trans* ratio than its predecessor and better reflects the ratios found in commercially available Tetra products, use of ICS-T2 will in most cases lead to a truer result than that obtained using ICS-T1.

### How to Purchase

Stocks of ICS-I2 and ICS-T2 have now been divided between ASBC (in USA) and Labor Veritas (in Switzerland), from which sources analysts can purchase 250 mg vials of each standard.\* Orders are dispatched by express mail to minimize risk of damage in transit.

Detailed information pertaining to each standard, including full instructions for use, is available on request and is automatically supplied with each purchase. For analysts requiring the DCHA-Rho and DCHA-Hexa standards, ICS-R1 and ICS-H1, these standards continue to be available as before. However, ICS-I1 and ICS-T1 will now be available only while stocks last.

RJHW

22nd September 2004

\*Purchasers in the USA, Canada, Central and South America should contact ASBC headquarters (email: [asbc@scisoc.org](mailto:asbc@scisoc.org); tel: +1 (651) 454-7250), while those in Europe and Africa should direct enquiries to Labor Veritas, Zürich, Switzerland (email: [admin@laborveritas.ch](mailto:admin@laborveritas.ch); tel: +41 (0) 44 283 29 30). Persons ordering from other parts of the world may make their approach to either party.