CERTIFICATE OF ANALYSIS

trans-ZEATIN-O-GLUCURONIDE (tZOGN)
$C_{16}H_{21}N_5O_7$, MW = 395.38

OLCHEMIM CODE: 001 597n
OLCHEMIM BATCH: ZOGN-A
MANUFACTURING DATE: August 2024
EXPIRATION DATE: August 2034
CAS: 147663-51-2
BATCH SIZE: 0.5 g

SYNTHESIS
Purified by reverse-phase chromatography and re-crystallised from methanol. A colourless, amorphous solid, sparingly soluble in water or methanol. Best solvent dimethyl sulfoxide.

ANALYSIS
MELTING POINT: 176.5 – 178°C (decomposition).
UV: Determined after drying to constant weight over phosphorus pentoxide in vacuum.

A max aq. ethanol	276 nm, 270 nm (shoulder), 283 nm (shoulder)
aq. ethanol/HCl	279 nm
aq. ethanol/ammonia	276 nm, 270 nm (shoulder), 283 nm (shoulder)

TLC (silica - nBuOH/AcOH/water. 12/3/5). Single diffuse spot, Rf 0.16. No impurities detectable at approximate 50 µg loading. No tZ (0.66), 6-chloropurine (0.70). Purity 97+%
HPLC: Samples were dissolved in mobile phase (initial conditions) and separated on RP-column (150 mm □ 4.6 mm, 5µm) (Microsorb C18; Varian). The elution was performed with a methanolic gradient. The flow rate was 0.6 ml/min. The UV absorbance was scanned between 240 – 300 nm; purity 95+%.
MS: Data were collected in ESI (+) mode from 50 - 1000 Da, matches the reference spectrum □M+H] + = 396.3.

GUS hydrolysis: Zeatin-O-β-D-glucuronic acid, Na Salt (1 mg) was incubated with bacterial glucuronidase (Sigma type G-8271) in 50 mM sodium phosphate buffer at pH 7.0 at 37°C for 4 h. TLC (silica - chloroform/MeOH, 1/1) of 50 µg showed complete conversion to *trans*-zeatin. The identity of *trans*-zeatin in the enzyme hydrolysate was confirmed by co-chromatography of a 1:1 mix of the hydrolysate and authentic *trans*-zeatin in 3 different chromatographic systems.

STORAGE & STABILITY: ACID LABILE! HYGROSCOPIC!
Store over silica gel in tightly sealed container at –18°C.
NOTE: For quantitative work, therefore, calculate concentration from UV absorption.

FOR LABORATORY USE BY EXPERIENCED PERSONNEL ONLY; NOT FOR DRUG, HOUSEHOLD OR OTHER USES

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