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## ATODLEN 2

Rheoliad 3

Diwygiadau i'r Atodiad i Reoliad (EU) Rhif 231/2012 sy'n ymwneud â'r fanyleb ar gyfer E 960a glycosidau stefiol o Stevia (E 960 glycosidau stefiol yn flaenorol) ac i ychwanegu manyleb ar gyfer E 960c rebaudiosid M a gynhyrchir drwy addasu glycosidau stefiol o Stevia ag ensymau

**1.** Yn Rheoliad y Comisiwn (EU) Rhif 231/2012, mae'r Atodiad (manylebau ar gyfer ychwanegion bwyd gan gynnwys lliwiau a melysyddion a restrir yn Atodiadau 2 a 3 i Reoliad (EC) Rhif 1333/2008) wedi ei ddiwygio fel a ganlyn.

2. Yn lle pennawd y cofnod ar gyfer "E 960 STEVIOL GLYCOSIDES" rhodder-

## "E 960a STEVIOL GLYCOSIDES FROM STEVIA"

3. Yn y lle priodol, mewnosoder y cofnod a ganlyn-

## **"E 960c REBAUDIOSIDE M PRODUCED VIA ENZYME MODIFICATION OF STEVIOL GLYCOSIDES FROM STEVIA**

Synonyms			
Definition	Rebaudioside M is a steviol glycoside composed predominantly of rebaudioside M with minor amounts of other steviol glycosides such as rebaudioside A, rebaudioside B, rebaudioside D, rebaudioside I, and stevioside.		
	Rebaudioside M is obtained via enzymatic bioconversion of purified steviol glycoside leaf extracts (95% steviol glycosides) of the <i>Stevia</i> <i>rebaudiana</i> Bertoni plant using UDP-glucosyltransferase and sucrose synthase enzymes produced by the genetically modified yeasts <i>K</i> . <i>phaffî</i> (formerly known as <i>Pichia pastoris</i> ) UGT-a and <i>K. phaffî</i> UGT-b that facilitate the transfer of glucose from sucrose and UDP- glucose to steviol glycosides via glycosidic bonds. After removal of the enzymes by solid-liquid separation and heat treatment, the purification involves concentration of the rebaudioside M by resin adsorption, followed by recrystallisation of rebaudioside M resulting in a final product containing not less than 95% of rebaudioside M. Viable cells or the DNA of the yeasts <i>K. phaffî</i> UGT-a or <i>K. phaffî</i> UGT-b must not be detected in the food additive.		
Chemical name	Rebaudioside M: $13-[(2-O-\beta-D-glucopyranosyl-3-O-\beta-D-glucopyranosyl-\beta-D-glucopyranosyl)oxy]kaur-16-en-18-oic acid, 2-O-\beta-D-glucopyranosyl-3-O-\beta-D-glucopyranosyl-\beta-D-glucopyranosyl ester$		
Molecular formula	Trivial name	Formula	Conversion factor
	Rebaudioside M	C <sub>56</sub> H <sub>90</sub> O <sub>33</sub>	0.25
Molecular weight and CAS number	Trivial name	CAS Number	Molecular weight (g/mol)
	Rebaudioside M	1220616-44-3	1291.29
Assay	Not less than 95% rebaudioside M on the dried basis		

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Description	White to light yellow powder, approximately between 200 and 350 times sweeter than sucrose (at 5% sucrose equivalency)	
Identification		
Solubility	Freely soluble to slightly soluble in water	
рН	Between 4.5 and 7.0 (1 in 100 solution)	
Purity		
Total ash	Not more than 1%	
Loss on drying	Not more than 6% (105°C, 2h)	
Residual solvent	Not more than 5000 mg/kg ethanol	
Arsenic	Not more than 0.015 mg/kg	
Lead	Not more than 0.2 mg/kg	
Cadmium	Not more than 0.015 mg/kg	
Mercury	Not more than 0.07 mg/kg	
Residual protein	Not more than 5 mg/kg	
Particle size	Not less than 74 $\mu$ m (using a mesh #200 sieve with a particle size limit of 74 $\mu$ m)"	