

Nourseothricin (NTC) – gold standard lab antibiotic

(1) NTC for pro- and eukaryotes

Nourseothricin (NTC, sometimes also termed clonNAT) is a superior selection antibiotic for genetic modification of unicellular and complex organisms including bacteria, yeast, filamentous fungi, protozoa, microalgae as well as for manipulating plant and mammalian cells (ask for info) cells (ask for info)

NTC selection pressure is used for gene disruption, knock-out, complementation, replacement and overexpression, and the number of NTC engineered organisms (ask for info) is growing steadily.

For example, recently the

- human pathogenic fungus Coccidioides posadasii,
- wood-decay fungus Physisporinus vitreus,
- red yeasts Rhodosporidium kratochvilovae and Rhodotorula graminis,
- plant root colonizing biocontrol fungus Trichoderma atroviride,
- plant pathogenic fungus Verticillium dahlia,
- carotenoid producer Xanthophyllomyces dendrorhous,
- osmotolerant yeast Zygosaccharomyces rouxii,

• protozoon *Trypanosoma vivax*.

were reported to be NTC susceptible (Wise et al. 2013; Schubert et al. 2013, Abbott et al. 2013, Dubey et al. 2013, Santhanam et al. 2013, Gassel et al. 2014; Watanabe et al. 2013, Goyard et al. 2014)

(2) NTC mechanism

Figure 1: Chemical structure of Nourseothricin. The antibiotic consists of 4 components (C, D, E and F) differing in the number of β-lysine units (n = 1 to 4). The contents of most active D + F > 85 %.

NTC is an antibiotic of the streptothricin group produced by Streptomyces noursei. The antibiotic effect is based on inhibition of protein biosynthesis and induction of miscoding.

The NTC resistance is encoded by the bacterial sat1 or Streptomyces nat1 marker genes. Its codon usages and expression signals were adapted to various recipient hosts. The product of the resistance genes - Nouresothricin N-acetyltransferase - inactivates NTC by monoacetylation of the β -amino group of its β -lysine residue (Figure 1).

NTC is highly soluble in water and long-term stable as powder or in solution. It shows no or very low background, no cross-reactivity with other aminoglycoside antibiotics such as Hygromycin or Geneticin and no cross-resistance with therapeutic antibiotics.

Further, NTC is used for our eukaryotic expression system LEXSY.

References:

Abbott et al. (2013) Overcoming recalcitrant transformation and gene manipulation in *Pucciniomycotina* yeasts. *Applied Microbiology and Biotechnology* **97**:283-295.

Dubey et al. (2013) Role of the methylcitrate cycle in growth, antagonism and induction of systemic defence responses in the fungal biocontrol agent *Trichoderma atroviride*. *Microbiology* **159**:2492-2500.

Gassel *et al.* (2014) Genetic engineering of the complete carotenoid pathway towards enhanced astaxanthin formation in *Xanthophyllomyces dendrorhous* starting from a high-yield mutant. *Applied Microbiology and Biotechnology* **98**:345-350.

Goyard et al. (2014) In vivo imaging of trypanosomes for a better assessment of host-parasite relationships and drug efficacy. Parasitology International 63:260-268.

Santhanam et al. (2013) Verticillium dahliae Sge1 Differentially Regulates Expression of Candidate Effector Genes. Mol. Plant Microbe Interact. 26:249-256.

Schubert et al. (2013) Agrobacterium-mediated transformation of the white-rot fungus *Physisporinus vitreus Journal of Microbiological Methods* **95**:251-252.

Watanabe et al. (2013) Adaptation of the Osmotolerant Yeast Zygosaccharomyces rouxii to an Osmotic Environment Through Copy Number Amplification of FLO11D. Genetics 195:393-405.

Wise et al. (2013) Extracellular ammonia at sites of pulmonary infection with *Coccidioides posadasii* contributes to severity of the respiratory disease. *Microbial Pathogenesis* **59-60**:19-22.

NTC products

LEXSY NTC (Nourseothricin) is the prime selection antibiotic for recombinant LEXSY strains. It can efficiently be used also for other host systems as Gram-positive and Gram-negative bacteria, Streptomycetes, yeast, filamentous fungi, protozoa, plants etc.

Product	Cat. No.	Amount	Cat. No.	Amount	Technical
					sheet
LEXSY NTC (Nourseothricin),	AB-101S	1 ml	AB-101L	5 ml	<u>OnLine</u>
sterile ready-to-go stock solution, 100 mg/ml	AB-101-10ML	10 ml	AB-101-50ML	50 ml	
LEXSY NTC (Nourseothricin), powder (non-sterile)	AB-102L	1 g	AB-102XL	5 g	<u>OnLine</u>
	AB-102-25G	25 g	AB-102-100G	100 g	
LEXSY Bleo , sterile ready-to-go stock solution, 100 mg/ml	AB-103S	1 ml	AB-103L	5 ml	<u>OnLine</u>
LEXSY Neo, sterile ready-to-go stock solution, 50 mg/ml	AB-105S	1 ml	AB-105L	5 ml	<u>OnLine</u>
LEXSY Tet , sterile ready-to-go stock solution, 10 mg/ml	AB-106S	1 ml	AB-106L	5 ml	<u>OnLine</u>
(for inducible protein expression in LEXSY host T7-TR)	AB-106XL	10 ml	AB-106XXL	50 ml	

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