Interim pharmacokinetic and pharmacodynamic data from the first-in-human study of NUC-3373, a pyrimidine nucleotide analogue, in patients with advanced solid tumours

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BACKGROUND

- Key cancer resistance mechanisms linked to reduced efficacy, poor prognosis and off-target toxicity with a 5-fluorouracil (5-FU) regimen[1]
- Poor PK properties of 5-FU often necessitate prolonged administration times (e.g. 46 hours)
- Fluorodeoxyuridine-monophosphate (FUDR-MP) is the main anti-cancer metabolite of 5-FU, which binds to and inhibits thymidylate synthase (TS), leading to cancer cell death

5-FU Resistance Mechanisms

Susceptibility to breakdown

- Over 85% of 5-FU broken down by dihydropyrimidine dehydrogenase (DPD)[2]
- Thymidine phosphorylase (TP), commonly overexpressed in tumours[1] or introduced by mycoplasma infection[3], also breaks down fluoropyrimidines
- Metabolic degradation results in generation of toxic metabolites, such as dihydrofluorouracil (dhFU) and α-fluoro-β-alanine (FBAL)[4]

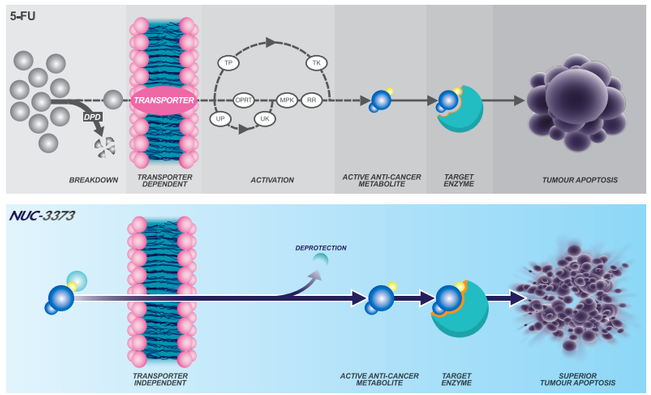
Requirement of activation

- 5-FU is a pro-drug that requires complex intracellular enzymatic activation to generate FUDR-MP[1]
- Deficient enzymatic activation linked to poor prognosis

Reliance on active transport

- Low expression of the nucleoside transporter hENT1 is associated with 5-FU resistance[5]

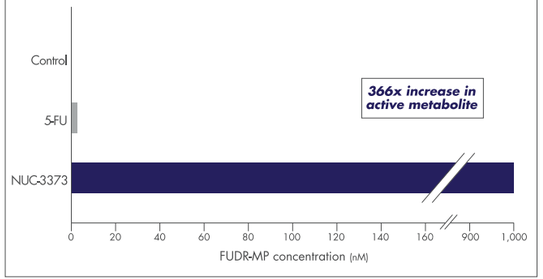
NUC-3373 and 5-FU mechanism of action



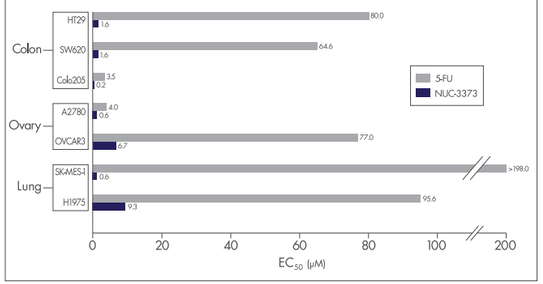
NUC-3373

- A pyrimidine nucleotide analogue designed to overcome key cancer resistance mechanisms associated with 5-FU[6,7]
- A phosphoramidate of FUDR
- Generates 366× higher intracellular levels of FUDR-MP than 5-FU *in vitro*
- Up to 330× significantly greater cytotoxicity *in vitro* than 5-FU
- Significantly greater anti-cancer activity *in vivo* compared to 5-FU
- Not degraded by DPD, unlike 5-FU[8]
- Favourable toxicology profile compared to 5-FU

NUC-3373 generates significantly higher levels of intracellular FUDR-MP in HT29 human colorectal cancer cell line compared with 5-FU



Comparative anti-cancer effect of NUC-3373 and 5-FU in human colorectal, ovarian and lung cancer cell lines



METHODS

- NUC-3373 administered as a short IV infusion on days 1, 8, 15 and 22 of a 28-day cycle in the ongoing NuTide:301 study
- The first 4 patient cohorts received NUC-3373 at 125 mg/m², 250 mg/m², 500 mg/m² and 750 mg/m²
  - These doses of NUC-3373 are equivalent to 27-160 mg/m² of 5-FU, well below the usual IV bolus dose range (400-500 mg/m²) and significantly below the continuous infusion dose range (2,000-3,000 mg/m²)[9]
- Blood collected pre-dose and at 11 time-points up to 48 hours post-dose during Cycle 1
- Plasma and intracellular metabolites measured by UPLC-MS/MS; western blotting of extracted PBMCs measured TS within ternary complexes (TS-T)

Primary objective

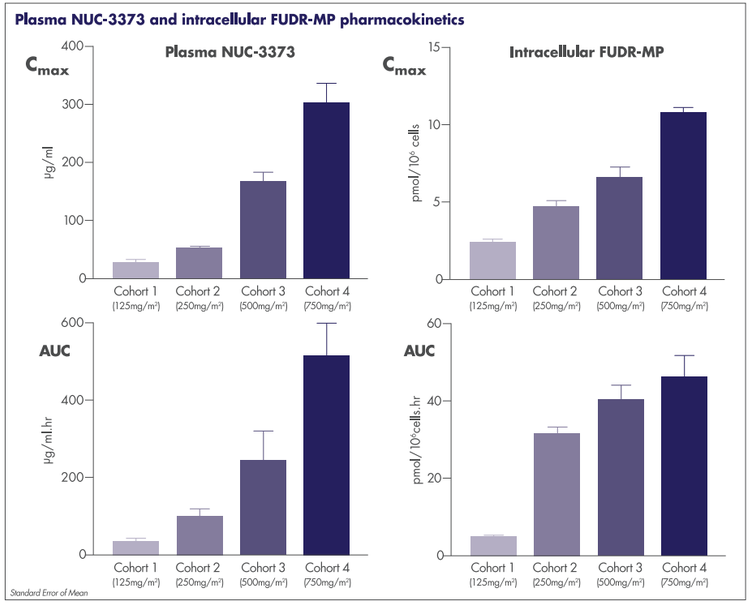
- Establish the recommended Phase II dose (RP2D)

Secondary objective

- Safety, PK/PD and anti-tumour activity

RESULTS

Plasma NUC-3373 and intracellular FUDR-MP pharmacokinetics



- 21 patients dosed in the first 4 dose cohorts, median age 57 years (range 20-77)
- 10 primary cancer types, the majority (57%) being colorectal cancer
- Mean C_{max} and AUC were dose proportional and reproducible
- Linear PK was confirmed across the studied dose range with clearance of 2.5±0.30 L/h and plasma $t_{1/2}$ of 9.7±0.82 hours
- Intracellular FUDR-MP detectable at 5 minutes post-infusion with $t_{1/2}$ of 14.9±1.44 hours and still present at 48 hours
- Mean intracellular C_{max} and AUC_{0-24} of FUDR-MP in the 500 mg/m² cohort were 6.5 pmol/10⁶ cells and 39.9 pmol/10⁶ cells/h, and in the 750 mg/m² cohort were 10.7 pmol/10⁶ cells and 43.6 pmol/10⁶ cells/h
- Within 1 hour of infusion, FUDR-MP was present within TS-T leading to depletion of the intracellular dTMP pool within 2-4 hours
- Toxic metabolites dhFU and FBAL were undetectable intracellularly or in plasma

NUC-3373 PK profile comparison with 5-FU

	NUC-3373	5-FU
Plasma half-life	9.7 hours	8-14 minutes
FUDR-MP (in PBMCs)	Detected (dose proportional)	Undetected[10]
Thymidylate Synthase inhibition	Strong	Weak
Intracellular levels of dTMP	Depleted	No change
Toxic metabolites (dhFU, FBAL)	Undetected	High levels

CONCLUSION

- NUC-3373 is a novel pyrimidine nucleotide analogue that overcomes the key cancer resistance mechanisms associated with 5-FU, and its other forms, floxuridine and capecitabine
- NUC-3373 generates very high intracellular concentrations of the active anti-cancer metabolite, FUDR-MP
- TS is efficiently inhibited and sequestered into TS-T, depleting the pool of dTMP within 2-4 hours
- Lack of toxic metabolite generation, suggestive of an improved tolerability profile compared to 5-FU
- NUC-3373 is resistant to DPD-mediated metabolism unlike 5-FU
- NUC-3373 has an advantageous PK/PD profile compared to 5-FU, which may allow for a more convenient dosing regimen, favourable safety profile and enhanced efficacy
- Dose escalation is ongoing to establish the RP2D

1. Longley DB *et al*, 2003. 2. Diasio RB & Harris BE, 1999. 3. Huang S *et al*, 2001. 4. Deboever G *et al*, 2013. 5. Tsujie M *et al*, 2007. 6. McGuigan C *et al*, 2011. 7. Vande Voorde J *et al*, 2011. 8. Blagden *et al*, 2016. 9. FOLFOX6, FOLFIRI, FUFOX and IFL regimens. 10. Derissen *et al*, 2016.

Data current as of 1 Sept 2017. Data cleaning ongoing.