Title of study:	Pilot study to assess the diagnostic value of the cholyl lysyl fluorescein (NRL972) assay in cystic fibrosis patients with known advanced liver disease.
Investigator:	Dr David Honeybourne, MD MSc FRCP
Study centre:	Birmingham, UK.
Publication (reference):	None.
Dates of study:	24 November 2003 to 05 November 2004.
Clinical phase:	Phase II.
Objectives:	To establish the value of NRL972 in cystic fibrosis patients with known advanced liver disease (CFLD). To assess the potential use of NRL972 in the monitoring of cystic fibrosis patients for liver diseases. To correlate the NRL972 functional test to the results of liver imaging obtained by ultrasound. To compare the use of the NRL972 test to clinical features and biochemical parameters assessing liver function. To investigate the safety of the use of NRL972 in cystic fibrosis patients with advanced cholestatic liver disease.
Primary efficacy variable: Secondary efficacy variables:	Determination of NRL972 clearance to establish the diagnostic usefulness of the NRL972 test in the management of cystic fibrosis patients with known cholestatic, focal liver disease. Correlation of NRL972 values with: • ultrasound findings of the liver • biochemical parameters (e.g. liver enzymes, bilirubin, albumin, clotting factors) • the clinical status of the patient.
Methodology:	This was a single-centre, open-label, non-randomised, pilot study in male and female adult patients with cystic fibrosis and known advanced liver disease (by clinical judgement). Each patient gave written informed consent before undergoing a screening clinical examination and assessment of entry criteria. Eligible patients were scheduled to have two NRL972 tests, separated by at least 2 weeks. An ultrasound examination was to be performed if patients had not had one in the previous 18 months.
Number of patients:	Planned: 12 Actual entered: 9 Analysed: 9 (per protocol set and safety set) The study was terminated early because of slow recruitment.
Diagnosis and main inclusion criteria:	Male or female patients with cystic fibrosis without acute exacerbation of lung infection; aged 18 years or over; presence of advanced liver disease with a splenomegaly (size >12.5 cm) and irregular and/or nodular appearance of liver as determined by ultrasound (clinical judgement).
Dosage and administration:	Test product: NRL972 solution (2 mg in 5 mL water for injection) administered by intravenous injection into antecubital vein over 15 seconds.

	Batch number: 02I02.3
Duration of treatment:	The study drug was administered to each patient on two separate occasions (single doses) separated by a period of at least 2 weeks (14 days)
Criteria for evaluation:	Bioanalytical: Blood samples were taken at pre-defined time points up to 60 minutes after each injection of NRL972. NRL972 concentrations in plasma were measured using a validated high performance liquid chromatography (HPLC) method over the concentration range of 10 to 1500 ng/mL. Urine samples were collected for the period 0-3 hours post-dose. NRL972 concentrations in urine were measured using a validated HPLC method over the range of 10 to 500 ng/mL. Efficacy: NRL972 clearance measurements were calculated: area under the plasma concentration versus time curve from 0-60 minutes (AUC(0-60)); clearance (CL) from 0-60 minutes; CL using 2-point analysis for 30-10 minute and 30-15 minute pairings; CL ratios for 30/10, 30/15, 45/10 and 45/15 minutes; gradients of the slopes for NRL972 concentrations. Pharmacokinetic: Maximum plasma concentration (C_{max}); time to C_{max} (t_{max}); half-life ($t_{½}$). Safety: Adverse events.
Statistical Methods:	Primary: Using a range of cut-offs based on the 95% confidence interval (CI) of the mean, standard deviation (SD) and standard error of the mean (SEM), the NRL972 clearance measurements were classified into 'reduced clearance' or 'normal clearance' and the sensitivity of this diagnosis was compared with the reference diagnosis. Patients were then categorised as 'true positive diagnosis' or 'true negative diagnosis'. Secondary: NRL972 clearance measurements were correlated with individual ultrasound findings of the liver, biochemical parameters, and clinical status.
Summary and Conclusions:	The diagnostic method based on the NRL972 clearance currently has neither a consensus of a possible 'cut-off' point or range of clearance values to classify NRL972 clearance into two categories of diagnosis: 'reduced clearance' (positive diagnosis) versus 'normal clearance' (negative diagnosis) nor a method to determine it automatically. Using a range of cut-offs, NRL972 clearance measurements were classified into 'reduced clearance' or 'normal clearance' and the sensitivity of this diagnosis was compared to the reference diagnosis. For patients with values at both test days the mean value was used for defining the cut-offs. Patients were then categorised either as 'True positive diagnosis' or 'False negative diagnosis'. Some strong correlations were observed between NRL972 AUC(0-60) and other parameters of interest, such as biochemical parameters and Westaby score of ultrasound findings. Reproducibility of the clearance measurements between Test Days 1 and 2 was generally good. There were no safety concerns following the administration of an injection of NRL972 (2 mg in 5 mL water for injection) in adult male and female CFLD patients. The injection was well tolerated in the patients who were considered to be unwell, having multiple organ impairment related to the underlying cystic fibrosis conditions.
Date of report:	09 March 2007

Norgine discontinued the development of NRL972 in July 2013.