Title:
A single-centre, open, controlled, randomised cross-over study in healthy male and female volunteers to investigate the effects of the injection volume and injection time on the pharmacokinetics of cholyl-lysyl-fluorescein (NRL972).

NORGINE Study №: NRL972-06/2007 (VOLT)

Investigators:

E Peterfai

Study centre(s):

Balatonfüred, Hungary

Publication (reference):

n.a.

Study period:	**Clinical Phase:** I
18 Mar 2008 (screening of 1st subject) – 29 May 2008 (end-of-trial in last subject)	

GCP-compliance:

The study was planned, conducted, analysed and reported in accordance with the pertinent GCP-Guidelines.

Objectives of the study:

To assess the effects of injection rate (part-A) and injection volume (part-B) on the plasma pharmacokinetics of NRL972 after a 2 mg iv injection of NRL972. Additionally, to evaluate the safety and tolerability of the medication under these conditions.

Study design:

Single-centre, controlled, randomised, open, four-period within-subject cross-over study with single iv injections of 2 mg NRL972 on four occasions with different injection rates (part-A: 5 mL in 5, 15, 30 and 60 sec) and injection volumes (part-B: 3, 5, 10, and 20 mL in 15 sec). Individual treatment days were to be at least one week apart for washout purposes. Part-A and -B could be conducted in parallel.

Number of subjects:

Twenty-four healthy volunteer subjects (twelve males, twelve females) in total; two groups of 12 subjects (six males, six females each).

Diagnosis and criteria for inclusion:

Male and female (of non-childbearing potential or while taking medically appropriate contraception), Caucasian subjects, 21 – 45 years of age, Body Mass Index (BMI) of 20 to 28 $kg.m^{-2}$ and body weight (BW) of 50 to 100 kg who were confirmed to be healthy on the basis of extensive screening investigation (medical history, physical examination, vital functions, 12-lead ECG, clinical laboratory safety tests [haematology, clinical chemistry, urinalysis, serology, screening tests for substances of abuse and alcohol]), and who were able and willing to provide informed consent.

Test product, dose, batch N°:

Cholyl-L-lysine-fluorescein (synonyms: CLF, cholyl-lysyl-fluorescein; INN Fluorescein lisicol tri-sodium salt; Development Code: NRL972), NORGINE Ltd., solution for iv injection (2mg NRL972 in solution for injection), 2 mg administered once by iv injection on four occasions in each subject with different rates (subjects in part-A) or different volumes of injection (subjects in part-B). The standard reference for the administration of the 2 mg dose is the 5 mL volume, injected over 15 seconds.

IMP containing NRL972 in two concentrations was used:
2 mg / 5 mL of injection: IMP Batch-N°: NOR-p004 (all treatments of part-A, treatments B-R, B-T2 [diluted to 10 mL], B-T3 [diluted to 20 mL] of part-B)
2 mg / 3 mL of injection: IMP Batch-N°: NORp007 (treatment B-T1 of part-B)

Reference product, batch N°:

Not applicable

Duration of treatments:

One single administration of 2 mg NRL972 on the morning of Day D01 on four periods at least 1 week apart

Schedule:

- Screening (SCR) visit: Eligibility assessment (EA): between 14 and 2 days before admission for the first period
- Four periods at least one week apart, with a single dose of 2 mg NRL972 administered at different rates each time (part-A) and using different volumes (part-B); for each period, the subjects were to be hospitalised from the evening before until 8:00 hours after injection of NRL972
- Wash-out interval: at least 1 week between consecutive treatment days D01 of four periods
- End-of-trial (EOT): within one week after NRL972 in the last period

CLINICAL PHARMACOLOGY FINDINGS

Subject disposition:

- Forty-four (44) subjects were screened for enrolment. Nineteen (19) subjects were not enrolled (reasons listed in Section 10.1).
- Twenty-five (25) subjects were enrolled. All enrolled subjects were randomised for treatment on four occasions. One subject enrolled in part-A (Subject A112) withdrew consent after the first period; the subject was replaced by subject A113 who was assigned to the same treatment sequence as discontinued subject A112. All further subjects completed the study in accordance with the protocol
- All 25 subjects were included in the safety data-set; all subjects with pharmacokinetic data for at least one study period were included in the pharmacokinetic evaluation.

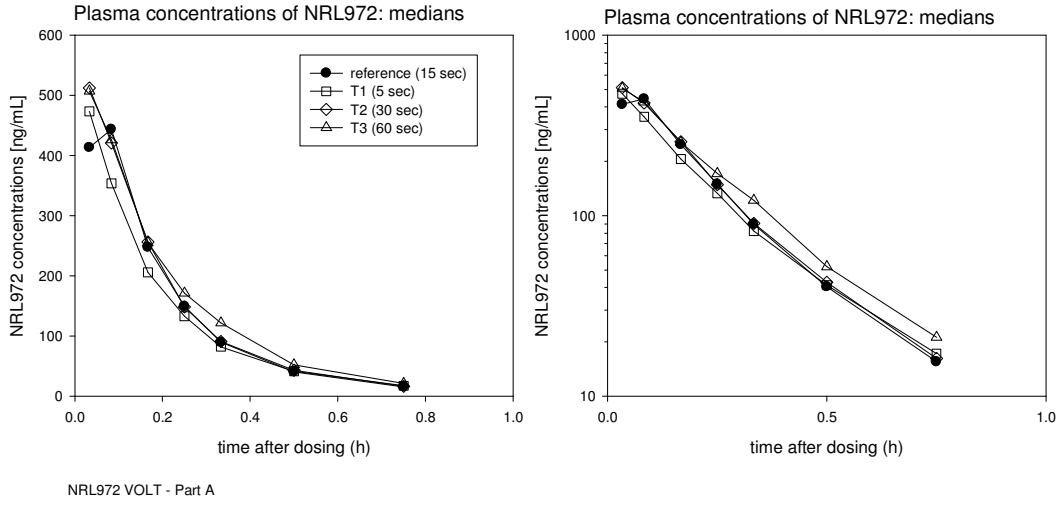
Demographics

All 25 randomised subjects (thirteen males, twelve females) were Caucasian: mean (± SD) age: 33.3 ± 5.8 years [range: 21 to 43]; mean body weight: 72.5 ± 12.7 kg [range: 52 to 98]; mean BMI: 24.9 ± 2.1 kg.m-2 [range: 20.3 to 27.8]. The demographic features of the 24 subjects who completed the study (twelve males, twelve females) were similar: mean (± SD) age: 33.4 ± 5.9 years [range: 21 to 43]; mean body weight: 71.9 ± 12.6 kg [range: 52 to 98]; mean BMI: 24.8 ± 2.1 kg.m-2 [range: 20.3 to 27.8].

All subjects were judged to be healthy upon in-depth evaluation at the screening visit.

Part-A – EFFECTS OF THE INJECTION RATE ON THE PHARMACOKINETICS OF NRL972

The time course of the observed median plasma NRL972 concentrations (left: linear axis; right: log-linear axis) for the four treatments after the iv injection of 2 mg/5 mL NRL972 (A-R: 15-second injection; A-T1: 5-second; A-T2: 30-second; A-T3: 60-second injection) are shown below:



Descriptive statistics: In the following tables, the descriptive statistics of the main pharmacokinetic variables are reported using aMX: arithmetic mean; aSD: arithmetic standard deviation; MIN: minimum observed value; MED: median; MAX: maximum observed value; gM: geometric mean; gCV: geometric coefficient of variation; the following variables are reported: ($C(t_2)$:$C(t_1)$-concentration ratios (1/1), back-extrapolated concentration at time zero (C0 [ng/mL]), maximum observed plasma concentration (Cmax [ng/mL]), time of occurrence of Cmax (tmax [min]), clearance (CL [mL/min]); BW-normalised clearance (CL/BW [mL/min per kg]), and log-linear half-life (t½ [min]) as derived by extensive "full" non-compartmental analysis (NCA-F), "short" analysis (NCA-S) and by 2-point analysis (2P-ANA) based on the C(10) & C(30) concentrations.
Comparative statistics: The ANOVA-estimates of the effects of the injection rate on the pharmacokinetics of NRL972 are detailed by means of the least square adjusted means per treatment and estimated difference (linear) or ratio (log-linear) of the true treatment means plus 95% CI [LL to UL] along with the residual coefficient of variation (CV-%).

Part-A: Control treatment - 15-second iv injection - Descriptive statistics:

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.179	0.036	0.176	0.20	0.132	0.174	0.243
2P-ANA	C(45):C(10)	0.069	0.017	0.067	0.25	0.045	0.069	0.097
2P-ANA	C(45):C(15)	0.112	0.025	0.110	0.23	0.077	0.117	0.150
2P-ANA	C0	635.5	118.4	625.5	0.19	433.7	646.3	893.2
2P-ANA	t½	8.08	0.95	8.03	0.12	6.84	7.93	9.81
2P-ANA	CL	283.2	62.3	276.0	0.25	158.2	298.6	366.7
2P-ANA	CL/BW	3.946	0.790	3.871	0.21	2.600	3.919	5.496
NCA-F	tmax	4	2	3	0.51	2	4	5
NCA-F	Cmax	489.8	105.0	479.7	0.21	353.8	475.0	696.3
NCA-F	t½	10.10	2.43	9.87	0.22	7.36	9.73	16.18
NCA-F	CL	311.1	67.3	303.7	0.24	180.0	318.0	395.5
NCA-F	CL/BW	4.328	0.795	4.260	0.19	3.028	4.274	5.769
NCA-S	t½	9.74	1.64	9.62	0.17	7.36	9.73	12.50
NCA-S	CL	311.3	66.8	304.1	0.24	183.0	318.0	395.5
NCA-S	CL/BW	4.332	0.789	4.266	0.19	3.028	4.274	5.769

Part-A: Test treatment T1 (5-second iv injection) - Descriptive statistics

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.191	0.062	0.182	0.32	0.110	0.182	0.310
2P-ANA	C(45):C(10)	0.091	0.043	0.083	0.43	0.056	0.078	0.181
2P-ANA	C(45):C(15)	0.136	0.053	0.129	0.35	0.090	0.124	0.257
2P-ANA	C0	538.1	121.1	524.8	0.24	329.0	557.4	758.6
2P-ANA	t½	8.43	1.72	8.29	0.20	6.29	8.14	11.84
2P-ANA	CL	328.2	74.3	318.8	0.27	162.4	353.9	423.5
2P-ANA	CL/BW	4.612	1.218	4.472	0.26	2.953	4.659	7.302
NCA-F	tmax	2	0	2	0.00	2	2	2
NCA-F	Cmax	481.1	112.5	469.2	0.24	290.7	473.5	727.9
NCA-F	t½	12.53	7.03	11.11	0.52	6.27	9.40	26.41
NCA-F	CL	343.7	81.3	332.5	0.29	155.6	366.1	451.9
NCA-F	CL/BW	4.824	1.316	4.664	0.28	2.830	4.945	7.791
NCA-S	t½	10.43	3.44	9.96	0.32	6.27	9.40	17.75
NCA-S	CL	345.8	79.3	335.2	0.28	160.7	366.1	451.9
NCA-S	CL/BW	4.855	1.294	4.702	0.27	2.922	4.945	7.791

Part-A: Test treatment T1 vs. Control - Comparative statistics

Method	Variable	R	T1	T1 - R	LL	UL	CV
2P-ANA	C(30):C(10)	0.179	0.191	0.012	-0.005	0.029	9.7
NCA-F	C0	635.5	538.1	-97.4	**-162.1**	**-32.8**	11.7
NCA-F	Cmax	489.8	481.1	-8.6	-59.4	42.2	11.1
NCA-F	CL	311.1	343.7	32.6	**6.5**	**58.6**	8.4
NCA-S	CL	311.3	345.8	34.4	**8.9**	**60.0**	8.3
2P-ANA	CL	283.2	328.2	45.0	**15.8**	**74.2**	10.1
NCA-F	CL/BW	4.33	4.82	0.50	**0.07**	**0.92**	9.9
NCA-S	CL/BW	4.33	4.86	0.52	**0.10**	**0.94**	9.7
2P-ANA	CL/BW	3.95	4.61	0.67	**0.20**	**1.13**	11.5
Method	**Variable**	**R**	**T1**	**T1 : R**	**LL**	**UL**	**CV**
NCA-F	t½	9.87	11.11	1.13	**0.92**	**1.38**	21.8
NCA-S	t½	9.62	9.96	1.04	**0.93**	**1.16**	11.9
2P-ANA	t½	8.03	8.29	1.03	**0.98**	**1.09**	5.4

In the above, contrasts reaching statistical significiance were shaded. The 5-second injection resulted in a slight increase in CL and CL/BW, whereas the estimated t½ tended to be longer. This reflects an increase in estimated distribution volume (also reflected by a lower C0) rather than a faster disposition rate (which would have been equivalent with a shorter t½).

Part-A: Test treatment T2 (30-second iv injection) - Descriptive statistics

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.189	0.055	0.181	0.30	0.121	0.175	0.275
2P-ANA	C(45):C(10)	0.078	0.025	0.074	0.35	0.046	0.075	0.115
2P-ANA	C(45):C(15)	0.116	0.027	0.113	0.24	0.081	0.116	0.154
2P-ANA	C0	636.1	125.3	623.4	0.22	371.6	655.6	812.4
2P-ANA	t½	8.35	1.48	8.23	0.18	6.56	7.95	10.75
2P-ANA	CL	279.9	74.6	270.1	0.29	158.8	286.8	391.4
2P-ANA	CL/BW	3.913	1.101	3.789	0.26	2.572	3.918	6.748
NCA-F	tmax	3	1	2	0.37	2	2	5
NCA-F	Cmax	509.9	95.5	502.0	0.18	370.8	512.1	725.1
NCA-F	t½	11.13	5.03	10.35	0.39	6.75	9.51	23.66
NCA-F	CL	303.4	82.6	291.6	0.31	158.2	331.4	416.1
NCA-F	CL/BW	4.242	1.225	4.090	0.29	2.591	4.336	7.175
NCA-S	t½	9.91	2.45	9.65	0.25	6.75	9.51	14.66
NCA-S	CL	304.1	81.5	292.7	0.31	163.1	331.4	416.1
NCA-S	CL/BW	4.253	1.211	4.106	0.28	2.630	4.336	7.175

Part-A: Test treatment T2 vs. Control - Comparative statistics

Method	Variable	R	T2	T2 - R	LL	UL	CV
2P-ANA	C(30):C(10)	0.179	0.189	0.009	-0.008	0.027	10.6
NCA-F	C0	635.5	636.1	0.6	-50.7	51.9	9.0
NCA-F	Cmax	489.8	509.9	20.1	-31.1	71.3	11.4
NCA-F	CL	311.1	303.4	-7.7	-32.0	16.6	8.8
NCA-S	CL	311.3	304.1	-7.3	-31.4	16.8	8.7
2P-ANA	CL	283.2	279.9	-3.3	-26.7	20.0	9.2
NCA-F	CL/BW	4.33	4.24	-0.09	-0.46	0.29	9.8
NCA-S	CL/BW	4.33	4.25	-0.08	-0.45	0.29	9.7
2P-ANA	CL/BW	3.95	3.91	-0.03	-0.39	0.33	10.2
Method	**Variable**	**R**	**T2**	**T2 : R**	**LL**	**UL**	**CV**
NCA-F	t½	9.87	10.35	1.05	0.92	1.20	14.6
NCA-S	t½	9.62	9.65	1.00	0.92	1.09	9.8
2P-ANA	t½	8.03	8.23	1.03	0.97	1.08	6.2

The 30-second injection had no effect on the pharmacokinetics of NRL972.

Part-A: Test treatment T3 (60-second iv injection) - Descriptive statistics

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.201	0.060	0.192	0.32	0.107	0.193	0.298
2P-ANA	C(45):C(10)	0.083	0.031	0.078	0.39	0.045	0.080	0.142
2P-ANA	C(45):C(15)	0.125	0.039	0.120	0.32	0.075	0.120	0.200
2P-ANA	C0	616.8	119.0	606.5	0.19	446.0	594.1	809.3
2P-ANA	t½	8.69	1.65	8.55	0.19	6.21	8.42	11.44
2P-ANA	CL	273.1	55.4	267.4	0.23	155.2	275.3	364.1
2P-ANA	CL/BW	3.858	1.000	3.751	0.25	2.773	3.681	5.913
NCA-F	tmax	3	1	3	0.48	2	2	5
NCA-F	Cmax	520.9	101.2	511.4	0.21	345.2	507.2	677.1
NCA-F	t½	11.99	6.51	10.96	0.42	6.01	10.67	31.44
NCA-F	CL	292.7	63.6	285.3	0.25	150.4	290.2	388.4
NCA-F	CL/BW	4.127	1.098	4.001	0.26	2.734	3.922	6.414
NCA-S	t½	10.71	2.78	10.37	0.28	6.01	10.67	16.10
NCA-S	CL	293.4	61.7	286.7	0.24	159.5	290.2	388.4
NCA-S	CL/BW	4.141	1.080	4.021	0.25	2.901	3.922	6.414

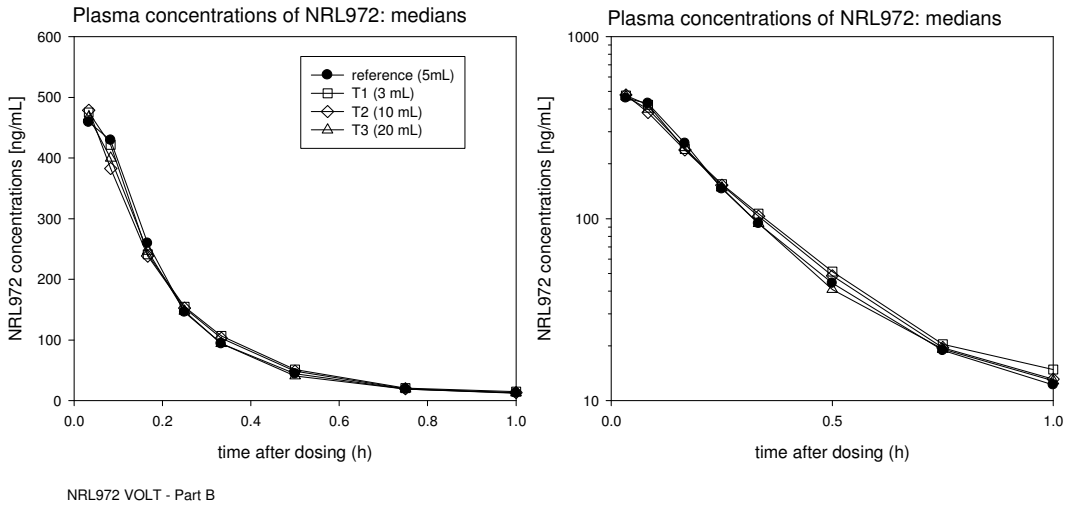
Part-A: Test treatment T3 vs. Control - Comparative statistics

Method	Variable	R	T3	T3 - R	LL	UL	CV
2P-ANA	C(30):C(10)	0.179	0.201	0.022	**0.000**	**0.044**	12.9
NCA-F	C0	635.5	616.8	-18.7	-67.5	30.1	8.7
NCA-F	Cmax	489.8	520.9	31.2	-35.1	97.5	14.6
NCA-F	CL	311.1	292.7	-18.4	-51.8	15.0	12.3
NCA-S	CL	311.3	293.4	-17.9	-51.3	15.4	12.3
2P-ANA	CL	283.2	273.1	-10.1	-38.1	17.9	11.2
NCA-F	CL/BW	4.33	4.13	-0.20	-0.63	0.23	11.2
NCA-S	CL/BW	4.33	4.14	-0.19	-0.61	0.23	11.1
2P-ANA	CL/BW	3.95	3.86	-0.09	-0.45	0.28	10.3
Method	**Variable**	**R**	**T3**	**T3 : R**	**LL**	**UL**	**CV**
NCA-F	t½	9.87	10.97	1.11	0.92	1.34	20.8
NCA-S	t½	9.62	10.37	1.08	0.92	1.26	17.2
2P-ANA	t½	8.03	8.55	1.07	0.99	1.14	7.7

The 60-second injection had no effect on the pharmacokinetics of NRL972 except for a slightly higher C(30):C(10)-ratio.

Part-B – EFFECTS OF THE INJECTION VOLUME ON THE PHARMACOKINETICS OF NRL972

The time course of the observed median plasma NRL972 concentrations (left: linear axis; right: log-linear axis) for the four treatments after the iv injection of 2 mg/5 mL NRL972 (B-R: 5 mL injection; B-T1: 3 mL injection; B-T2: 10 mL; B-T3: 20 mL are shown below:



Descriptive & comparative statistics: The descriptive and comparative statistics of the main pharmacokinetic variables are detailed as specified above for Part-A.

Part-B: Control treatment – 5 mL iv injection - Descriptive statistics:

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.194	0.071	0.182	0.40	0.104	0.194	0.327
2P-ANA	C(45):C(10)	0.083	0.037	0.075	0.50	0.037	0.082	0.155
2P-ANA	C(45):C(15)	0.127	0.041	0.121	0.33	0.070	0.124	0.204
2P-ANA	C0	628.3	151.4	610.3	0.26	376.6	640.6	819.9
2P-ANA	t½	8.52	1.96	8.32	0.23	6.12	8.46	12.41
2P-ANA	CL	283.7	76.9	273.1	0.31	137.8	284.9	392.8
2P-ANA	CL/BW	3.970	0.923	3.877	0.23	2.650	3.986	6.219
NCA-F	tmax	3	1	2	0.37	2	2	5
NCA-F	Cmax	510.3	100.5	502.1	0.19	417.3	470.2	717.6
NCA-F	t½	13.81	8.64	12.16	0.52	6.50	11.29	36.93
NCA-F	CL	303.3	83.1	290.7	0.33	130.0	314.6	431.8
NCA-F	CL/BW	4.268	1.167	4.128	0.28	2.499	4.056	6.854
NCA-S	t½	11.40	3.25	10.97	0.30	6.50	11.29	17.00
NCA-S	CL	305.0	81.6	293.2	0.32	139.1	320.2	431.8
NCA-S	CL/BW	4.294	1.138	4.163	0.26	2.675	4.056	6.854

Part-B: Test treatment T1 (3 mL iv injection) - Descriptive statistics

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.190	0.052	0.183	0.29	0.101	0.185	0.284
2P-ANA	C(45):C(10)	0.082	0.022	0.079	0.27	0.057	0.073	0.118
2P-ANA	C(45):C(15)	0.128	0.026	0.126	0.21	0.090	0.128	0.162
2P-ANA	C0	609.8	144.4	594.3	0.24	408.9	605.6	892.5
2P-ANA	t½	8.37	1.40	8.26	0.17	6.04	8.22	11.00
2P-ANA	CL	292.7	79.7	282.2	0.29	162.4	293.8	436.5
2P-ANA	CL/BW	4.108	0.941	4.008	0.24	2.631	3.948	5.513
NCA-F	tmax	2	1	2	0.27	2	2	5
NCA-F	Cmax	511.3	103.8	502.1	0.20	385.5	474.7	677.0
NCA-F	t½	11.30	3.88	10.72	0.35	6.34	10.15	18.84
NCA-F	CL	310.7	84.9	299.4	0.30	166.1	303.1	454.9
NCA-F	CL/BW	4.382	1.137	4.252	0.26	2.886	4.278	6.724
NCA-S	t½	10.85	3.18	10.43	0.30	6.34	10.15	16.06
NCA-S	CL	311.0	84.3	300.0	0.29	170.0	303.1	454.9
NCA-S	CL/BW	4.388	1.130	4.260	0.26	2.886	4.278	6.724

Part-B: Test treatment T1 vs. Control - Comparative statistics

Method	Variable	R	T1	T1 - R	LL	UL	CV
2P-ANA	C(30):C(10)	0.194	0.190	-0.004	-0.025	0.017	11.5
NCA-F	C0	628.3	609.8	-18.5	-95.2	58.2	13.2
NCA-F	Cmax	510.3	511.3	0.9	-65.8	67.7	13.9
NCA-F	CL	303.3	310.7	7.4	-31.7	46.5	13.5
NCA-S	CL	305.0	311.0	6.0	-34.1	46.1	13.8
2P-ANA	CL	283.7	292.7	9.0	-33.4	51.4	15.6
NCA-F	CL/BW	4.27	4.38	0.11	-0.48	0.71	14.7
NCA-S	CL/BW	4.29	4.39	0.09	-0.51	0.70	14.9
2P-ANA	CL/BW	3.97	4.11	0.14	-0.50	0.78	16.8
Method	**Variable**	**R**	**T1**	**T1 : R**	**LL**	**UL**	**CV**
NCA-F	t½	12.16	10.72	0.88	0.77	1.01	14.7
NCA-S	t½	10.97	10.43	0.95	0.85	1.06	11.7
2P-ANA	t½	8.32	8.26	0.99	0.93	1.06	7.3

The 3 mL injection had no effect on the pharmacokinetics of NRL972

Part-B: Test treatment T2 (10 mL iv injection) - Descriptive statistics

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.201	0.056	0.193	0.30	0.115	0.194	0.290
2P-ANA	C(45):C(10)	0.080	0.029	0.075	0.39	0.038	0.076	0.139
2P-ANA	C(45):C(15)	0.124	0.037	0.119	0.31	0.073	0.124	0.196
2P-ANA	C0	610.0	132.9	597.2	0.22	438.9	580.8	844.1
2P-ANA	t½	8.69	1.54	8.56	0.18	6.40	8.45	11.19
2P-ANA	CL	278.4	63.7	271.1	0.25	157.5	290.9	391.1
2P-ANA	CL/BW	3.888	0.584	3.850	0.15	3.029	3.768	5.117
NCA-F	tmax	3	1	2	0.37	2	2	5
NCA-F	Cmax	510.1	106.6	500.1	0.21	362.8	478.7	662.7
NCA-F	t½	13.03	6.75	11.89	0.44	8.20	10.14	29.14
NCA-F	CL	296.6	74.2	287.3	0.28	159.1	303.8	410.5
NCA-F	CL/BW	4.165	0.935	4.079	0.21	3.059	3.916	6.516
NCA-S	t½	11.08	2.69	10.80	0.24	8.20	10.14	16.45
NCA-S	CL	298.0	72.8	289.2	0.27	164.5	303.8	410.5
NCA-S	CL/BW	4.186	0.909	4.107	0.20	3.164	3.916	6.516

Part-B: Test treatment T2 vs. Control - Comparative statistics

Method	Variable	R	T2	T2 - R	LL	UL	CV
2P-ANA	C(30):C(10)	0.194	0.201	0.007	-0.015	0.029	12.4
NCA-F	C0	628.3	610.0	-18.3	-68.2	31.5	9.0
NCA-F	Cmax	510.3	510.1	-0.3	-48.4	47.9	10.5
NCA-F	CL	303.3	296.6	-6.7	-33.5	20.1	9.9
NCA-S	CL	305.0	298.0	-6.9	-33.5	19.6	9.8
2P-ANA	CL	283.7	278.4	-5.3	-30.1	19.6	9.8
NCA-F	CL/BW	4.27	4.17	-0.10	-0.50	0.30	10.5
NCA-S	CL/BW	4.29	4.19	-0.11	-0.50	0.29	10.3
2P-ANA	CL/BW	3.97	3.89	-0.08	-0.43	0.26	9.7
Method	**Variable**	**R**	**T2**	**T2 : R**	**LL**	**UL**	**CV**
NCA-F	t½	12.16	11.89	0.98	0.88	1.09	12.0
NCA-S	t½	10.97	10.80	0.99	0.92	1.06	7.8
2P-ANA	t½	8.32	8.56	1.03	0.96	1.11	8.1

The 10 mL injection had no effect on the pharmacokinetics of NRL972.

Part-B: Test treatment T3 (20 mL iv injection) - Descriptive statistics

Method	Variable	aMX	aSD	gM	gCV	MIN	MED	MAX
2P-ANA	C(30):C(10)	0.177	0.049	0.170	0.29	0.096	0.168	0.260
2P-ANA	C(45):C(10)	0.078	0.025	0.075	0.34	0.044	0.076	0.118
2P-ANA	C(45):C(15)	0.121	0.032	0.117	0.27	0.078	0.121	0.172
2P-ANA	C0	621.6	121.4	609.9	0.21	429.0	666.4	784.0
2P-ANA	t½	8.02	1.29	7.93	0.16	5.92	7.77	10.28
2P-ANA	CL	293.3	62.0	286.7	0.23	171.9	298.6	402.3
2P-ANA	CL/BW	4.156	0.946	4.072	0.21	3.158	3.908	6.595
NCA-F	tmax	3	1	2	0.37	2	2	5
NCA-F	Cmax	499.6	94.6	492.0	0.18	395.9	474.6	679.2
NCA-F	t½	11.76	6.23	10.73	0.43	7.08	9.76	29.27
NCA-F	CL	319.5	67.5	311.5	0.25	170.3	338.3	401.2
NCA-F	CL/BW	4.532	1.092	4.424	0.23	3.275	4.019	6.584
NCA-S	t½	10.48	3.10	10.08	0.29	7.08	9.76	15.48
NCA-S	CL	320.2	65.9	312.8	0.24	178.7	338.3	401.2
NCA-S	CL/BW	4.545	1.076	4.442	0.22	3.436	4.019	6.584

Part-B: Test treatment T3 vs. Control - Comparative statistics

Method	Variable	R	T3	T3 - R	LL	UL	CV
2P-ANA	C(30):C(10)	0.194	0.177	-0.017	-0.038	0.004	12.8
NCA-F	C0	628.3	621.6	-6.7	-86.8	73.4	14.3
NCA-F	Cmax	510.3	499.6	-10.8	-42.9	21.4	7.1
NCA-F	CL	303.3	319.5	16.2	-20.8	53.2	13.2
NCA-S	CL	305.0	320.2	15.2	-21.7	52.1	13.2
2P-ANA	CL	283.7	293.3	9.6	-31.9	51.1	16.0
NCA-F	CL/BW	4.27	4.53	0.26	-0.30	0.83	14.2
NCA-S	CL/BW	4.29	4.55	0.25	-0.31	0.81	14.1
2P-ANA	CL/BW	3.97	4.16	0.19	-0.46	0.83	17.7
Method	**Variable**	**R**	**T3**	**T3 : R**	**LL**	**UL**	**CV**
NCA-F	t½	12.16	10.73	0.88	**0.79**	**0.98**	12.1
NCA-S	t½	10.97	10.08	0.92	0.85	1.00	9.0
2P-ANA	t½	8.32	7.93	0.95	0.89	1.02	7.8

The 20 mL injection had no effect on the pharmacokinetics of NRL972 except for a slightly shorter t½ as estimated by NCA-F; there was a similar trend for NCA-S and 2P-ANA.

SAFETY

Wellbeing and adverse events (AE)

For each period, the subjects were hospitalised from the evening of D-1 until the evening of D01 at least 8:00 hours after administration of NRL972. During this time subjects were under close surveillance in the study clinic.

There were 2 AEs in 2/25 exposed subjects: moderate haematoma at the injection site after drawing the catheter for injection of the trial medication; diarrhoea with onset 6 days after last dosing; both were considered unrelated to the clinical trial medication. There were no SAEs; no AE led to premature discontinuation from the study.

Physical examination

A thorough physical examination took place at SCR and EOT. There were no relevant safety findings at the end of the study compared with the examination at SCR.

Vital functions

Recumbent blood pressure (SBP/DBP) and pulse rate (PR) were recorded at SCR and EOT and before each dosing with NRL972 and at scheduled times after each dosing. There was no indication of noteworthy average or individual SBP/DBP- or PR-findings over the course of the study.

12-lead ECG

A 12-lead digital ECG was recorded at SCR and EOT and at regular times before and after each NRL972 injection. There was no indication of relevant average or individual ECG-findings (HR, PQ, QT, QTc[Bazett], QTc[Fridericia], and QTc[Framingham]: untransformed and changes from baseline) over the course of the study.

CLINLAB: Clinical laboratory tests

Routine safety laboratory testing took place at SCR, prior to each NRL972 injection and at the EOT. Additionally, on the reference treatment days, blood for extensive clinical laboratory evaluation was taken at 0:30, 01:00, 2:00 and 4:00 hours after injection. The results were compared with the laboratory defined range of normality. Values outside the range of normality were highlighted and evaluated by the Investigators in terms of their clinical relevance. There were no safety-limiting or noteworthy values.

CONCLUSION

- Relative to the control treatment (R), the 30-second iv injection of NRL972 had no effect on the pharmacokinetics of NRL972. The 60-second iv injection of NRL972 had no effect on the pharmacokinetics of NRL972 but caused a marginal increase in C(30):C(10)-ratio (estimated μ_R: 0.179; μ_{T3}: 0.201; μ_{T3}-μ_R: 0.022, 95% CI: -0.000 to 0.044). The 5-second iv injection of NRL972 had a marginal effect on the pharmacokinetics of NRL972: there was no difference in C(30):C(10)-ratio (estimated μ_R: 0.179; μ_{T1}: 0.191; μ_{T1}-μ_R: 0.012, 95% CI: -0.005 to 0.029) and Cmax was about the same (estimated μ_R: 489.8 ng/mL; μ_{T1}: 481.1 ng/mL; μ_{T1}-μ_R: -8.6 ng/mL, 95% CI: -59.4 to 42.2). However, the back-extrapolated C0 was lower (estimated μ_R: 635.5 ng/mL; μ_{T1}: 538.1 ng/mL; μ_{T1}-μ_R: -97.4, 95% CI: -162 to -32.8) and the clearance CL [NCA-F] was slightly higher (estimated μ_R: 311.1 mL/min; μ_{T1}: 343.7 mL/min; μ_{T1}-μ_R: 32.6 mL/min, 95% CI: 6.5 to 85.6) also when normalised for body weight as CL/BW [NCA-F] (estimated μ_R: 4.33 mL/min per kg; μ_{T1}: 4.82 mL/min per kg; μ_{T1}-μ_R: 0.50 mL/min per kg, 95% CI: 0.07 to 0.92). In contrast, t½ [NCA-F] tended to be longer (estimated μ_R: 9.87 min; μ_{T1}: 11.11 min; μ_{T1}:μ_R: 1.13, 95% CI: 0.92 to 1.38). This finding is compatible with a slight increase in the estimated distribution volume when NRL972 is administered by a very fast iv injection. This difference is not likely to be of clinical relevance
- Relative to the control treatment (R), the 3 mL and 10 mL iv injections of NRL972 had no effect on the pharmacokinetics of NRL972. For the 20 mL injection, there was also no effect on the pharmacokinetics of NRL972 except for slightly shorter t½ [NCA-F] (estimated μ_R: 12.16 min; μ_{T3}: 10.73 min; μ_{T3}:μ_R: 0.88, 95% CI: 0.79 to 0.98). This small effect is unlikely to be of clinical relevance.
- Over the investigated range (injection rates of 5 to 60 seconds; injection volumes of 3 to 20 mL for a dose of 2 mg NRL972), effects of the injection rate or volume – if any – were very small and unlikely to be of clinical relevance, or of any significance when using the pharmacokinetics of NRL972 to differentiate and/or categorise biliary clearance dysfunction.
- The iv injections of 2 mg NRL972 on these four occasions per subject were very well tolerated; there were no relevant adverse events attributable to the investigational clinical trial medication. There were no noteworthy findings upon physical examination, for vital signs (recumbent blood pressure and pulse rate), and ECG (heart rate, atrio-ventricular and intra-cardiac conduction and QT/QTc) that had to be considered safety-limiting. There were no safety limiting noteworthy findings or changes in the clinical laboratory tests.