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1 **Hypothermic liquid ventilation prevents early hemodynamic dysfunction and**
2 **cardiovascular mortality after coronary artery occlusion**
3 **complicated by cardiac arrest in rabbits**

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28 **Key words:** Cardiac arrest – Myocardial infarction – Hypothermia – Resuscitation – Liquid
29 ventilation

30

31 **Abstract**

32 **Objective:** Ultrafast and whole-body cooling can be induced by total liquid ventilation (TLV)
33 with temperature-controlled perfluorocarbons. Our goal was to determine whether this can
34 afford maximal cardio- and neuroprotections through cooling rapidity when coronary
35 occlusion is complicated by cardiac arrest.

36 **Design:** Prospective, randomized animal study.

37 **Setting:** Academic research laboratory.

38 **Subjects:** Male New-Zealand rabbits.

39 **Interventions:** Chronically instrumented rabbits were submitted to coronary artery occlusion
40 and ventricular fibrillation. After 8-min of cardiac arrest, animals were resuscitated and
41 submitted to a normothermic follow-up (Control group) or to 3-h of mild hypothermia induced
42 by TLV (TLV group) or by combination of cold saline infusion and cold blankets application
43 (Saline group). Coronary reperfusion was permitted 40-min after the onset of occlusion. After
44 awakening, rabbits were followed during 7 days.

45 **Measurements and main results:** Ten animals were resuscitated in each group. In the
46 Control group, all animals secondarily died from cardiac/respiratory failure (8/10) or
47 neurological dysfunction (2/10). In the Saline group, the target temperature of 32°C was
48 achieved within 30-45 min after cooling initiation. This slightly reduced infarct size vs Control
49 ($41\pm 16\%$ vs $54\pm 8\%$ of risk zone, respectively; $p < 0.05$) but failed to significantly improve
50 cardiac output, neurological recovery and survival rate (3 survivors, 6 death from
51 cardiac/respiratory failure and 1 from neurological dysfunction). Conversely, the 32°C
52 temperature was achieved within 5-10 min in the TLV group. This led to a dramatic reduction
53 in infarct size ($13\pm 4\%$; $p < 0.05$ vs other groups) and improvements in cardiac output,
54 neurological recovery and survival (8 survivors, 2 deaths from cardiac/respiratory failure).

55 **Conclusions:** Achieving hypothermia rapidly is critical to improve the cardiovascular
56 outcome after cardiac arrest with underlying myocardial infarction.

57

58

59 **Introduction**

60 Out-of-hospital cardiac arrest is a leading cause of death in the world. After
61 cardiopulmonary resuscitation, comatose survivors require integrated post-resuscitation care
62 (1). Among the recommended treatments, hypothermia provides neuroprotection (2) while
63 percutaneous coronary interventions is intended for revascularization during coronary artery
64 occlusion (CAO) (3). Since hypothermia also decreases myocardial susceptibility to ischemia
65 (4-6), it could potentiate the cardioprotective effect of reperfusion therapies and further
66 improve hemodynamics when cardiac arrest is associated with coronary obstruction. This
67 cardiac benefit could however be compromised if hypothermia is only achieved into the
68 coronary reperfusion phase (4-6), supporting the importance of cooling the heart quickly.
69 This importance has also been evidenced for the neurological effect of hypothermia in animal
70 models of global (7-10) and brain (11) ischemia. Here, we hypothesize that rapidity of cooling
71 is also critical to achieve maximal protection and to prevent cardiovascular mortality when
72 cardiac arrest is combined with CAO.

73 In order to investigate the importance of the cooling rate, we previously studied ultra-
74 fast cooling through total liquid ventilation (TLV). It consists in the tidal ventilation of lungs
75 with temperature-controlled perfluorocarbons that can cool the body while maintaining gas
76 exchanges (12). After global ischemia, TLV provided a greater neurological benefit and
77 survival improvement than a conventional cooling in a rabbit model (12). The superiority of
78 TLV was also shown regarding infarct size reduction in rabbits submitted to CAO (5, 6, 13).
79 Importantly, these previous studies were conducted in pure models of cardiac arrest or CAO
80 but they did not reproduce the complex interactions occurring *in vivo* during their
81 combination, as often observed in patients (1, 3).

82 In the present study, we aimed to determine whether cooling rapidity, assessed by
83 comparison of TLV with a conventional "load-dependent" hypothermia (cold saline infusion
84 plus external cooling), could be actually critical to achieve maximal cardiac, neurological and
85 survival benefits after combined cardiac arrest and CAO in chronically instrumented rabbits.

86 **Materials and Methods**

87 The experiments were conducted in accordance with French official regulations, after
88 approval by the local ethical review board. They conformed to the “*Position of the American*
89 *Heart Association on research animal use*”.

90 Animal preparation

91 Male New-Zealand rabbits were anesthetized using zolazepam, tiletamine and
92 pentobarbital (all 20-30 mg/kg i.v.). They were chronically instrumented with a peri-coronary
93 pneumatic occluder, an aortic cardiac output probe (PS-Series Probes, Transonic, NY, USA)
94 and an electrode upon the chest.

95 Coronary occlusion, cardiac arrest and cardiopulmonary resuscitation

96 Two weeks after surgery, animals were reanesthetized, intubated and mechanically
97 ventilated ($FiO_2=100\%$; $V_t=10$ ml/kg; 26 breaths/min). The peri-coronary occluder was
98 inflated to induce CAO. Two minutes later, ventricular fibrillation was induced by passing an
99 alternating current (10 V, 4 mA) between the chest electrode and another electrode within
100 the esophagus. After 8 min of untreated fibrillation, cardiopulmonary resuscitation was
101 started using manual lateral chest compressions (~200 compressions/min), electric attempts
102 of defibrillation (5-10 J/kg) and intravenous administration of epinephrine (15 μ g/kg i.v.).
103 Mechanical ventilation was stopped throughout the cardiac arrest period and restarted at the
104 onset of cardiopulmonary resuscitation. Resumption of spontaneous circulation (ROSC) was
105 considered as an organized cardiac rhythm associated with a mean arterial pressure above
106 40 mmHg during at least 1 min. Coronary reperfusion was permitted through occluder
107 deflation after 40 min of CAO. After ROSC, administration of epinephrine was permitted
108 during 7 h to maintain mean arterial pressure at ~80 mmHg. Mechanical ventilation was
109 continued until awakening of the animals.

110

111 Experimental protocol

112 After ROSC, rabbits randomly underwent life support under normothermic conditions
113 (Control group) or with therapeutic hypothermia (TLV and Saline groups). In the TLV group,
114 hypothermia was induced by 20 min of liquid ventilation after filling the lungs with 10 ml/kg of
115 perfluorodecalin, as previously described (5, 6, 12, 13). The liquid ventilator was set initially
116 to a respiratory rate of 6 breaths/min and a tidal volume of ~7-10 ml/kg. If necessary,
117 ventilatory parameters were adjusted to maintain saturation of peripheral oxygen (SpO₂) over
118 90%. The temperature of perfluorocarbon was initially set at 20°C and progressively
119 increased to 32°C to maintain oesophageal temperatures at ~32°C. After 20 min of TLV,
120 animals resumed to conventional gas ventilation. In the Saline group, hypothermia was
121 induced by cold blankets over the skin and infusion of cold (4°C) saline (NaCl 0.9%, 30 ml/kg
122 i.v. over 30 min). In both Saline and TLV groups, hypothermia was maintained externally at
123 32°C during 3 h after the onset of the cooling protocol. Subsequently, animals were actively
124 rewarmed using thermal pads and infrared lights until weaning from conventional ventilation
125 and awakening.

126 Investigated parameters

127 Hemodynamic parameters were continuously recorded using external
128 electrocardiogram, blood pressure in the ear artery and cardiac output flowmeter (Flowmeter-
129 T206, Transonic, NY, USA).

130 Throughout the 7 days following cardiac arrest, the animals underwent a regular
131 survival and clinical follow-up. Neurological dysfunction was assessed using a previously
132 described score system (0-10% = normal; 100% = brain death) (12, 14). In accordance with
133 the ethical committee, the animals showing a neurological dysfunction score above 60%
134 during 48 hours were euthanized and considered as dead from neurological dysfunction. At
135 the end of the 7 days of follow-up, surviving rabbits were reanesthetized and a pressure
136 catheter (SciSense, London, Ontario, Canada) was introduced into the left ventricle for
137 measurement of end-diastolic pressure and left ventricular rate of pressure development.

138 These parameters were also measured in a group of Sham rabbits neither submitted to
139 cardiac arrest nor hypothermia.

140 Post-mortem analysis

141 Seven days after cardiac arrest, surviving animals were euthanized for organ
142 sampling. Pathological analyses were also conducted in the animals that died prematurely. In
143 the heart, risk zone and infarct sizes were determined using triphenyltetrazolium chloride
144 staining (5, 13). In all organs, the lesion severity was assessed by histology and quantified
145 using a 0-3 score system (12). For the brain, a neuropathologist semi-quantitatively and
146 blindly assessed the amount of ischemic neurons in the cortex, hippocampus and
147 cerebellum, as previously described (12). The brain score was the mean value obtained for
148 these three areas. For lungs, we assessed two separate scores for cardiogenic lesions
149 (serous edema and/or congestion) and infectious complication of bronchopneumonia,
150 respectively (12). In the myocardium, these analyses were performed in a remote area, out
151 of the risk zone.

152 Statistical analyses

153 Data were expressed as mean±SD. The normality of data distribution was tested
154 using the Kolmogorov-Smirnov analysis. For normally distributed parameters, comparisons
155 were made between groups using a one-way ANOVA at selected time-points as the number
156 of animals varied among time. Post-hoc analyses were performed using the Fisher Least
157 Significant Difference Method. For non-normally distributed parameters, neurological
158 dysfunction and histological scores, comparisons were made using a non-parametric
159 Kruskal-Wallis test followed by a Mann-Whitney analysis. Survival curves were obtained
160 using a Kaplan-Meier analysis and compared between group using a log-rank test.
161 Significant differences were determined at $P \leq 0.05$.

162

163 **Results**

164 Out of the 40 rabbits submitted to cardiac arrest, 30 animals were successfully
165 resuscitated and included in the final experimental groups (n=10 / group). The time to ROSC
166 was 2.9 ± 0.8 , 2.8 ± 1.4 and 3.1 ± 1.4 min in Control, TLV and Saline groups, respectively.
167 Temperatures decreased very rapidly after the onset of cooling in TLV as compared to Saline
168 group (Figure 2).

169 TLV prevents early hemodynamic dysfunction

170 As illustrated in Figure 3, TLV reduced left ventricular dysfunction after resuscitation.
171 After 30 min of CAO, cardiac output and stroke volumes were indeed increased in TLV vs
172 Control and Saline groups. Heart rate was conversely decreased throughout hypothermia in
173 both TLV and Saline groups (Table I). One day after cardiac arrest, cardiac output was again
174 increased in survivors of the TLV group as compared to Control and Saline groups.
175 Hypothermia also decreased the amount of epinephrine required to maintain mean arterial
176 pressure at ~ 80 mmHg after cardiac arrest (336 ± 209 , 305 ± 153 vs 751 ± 488 $\mu\text{g}/\text{kg}$ in TLV and
177 Saline vs Control groups, respectively; $p < 0.05$). Troponin I blood release was also
178 dramatically attenuated in TLV vs Control and Saline groups (Table II).

179 TLV improves cardiovascular outcome and provides cardioprotection

180 As shown in Figure 4 (Panels A-B), the hemodynamic improvements observed in the
181 TLV group was associated with a reduced mortality, in particular from cardiovascular and/or
182 respiratory failures. Seven days after cardiac arrest, the survival rate achieved 80% in TLV
183 group vs 0% and 30% in Control and Saline groups, respectively. As shown in Table III, left
184 ventricular performance was not altered in survivors of the TLV group as compared to Sham
185 animals, suggesting a complete recovery. Conversely, a pattern of heart failure was
186 observed in the 2 surviving rabbits of the Saline group.

187 Despite similar risk zone sizes in all groups (35 ± 7 , 34 ± 6 and 40 ± 6 % of the left
188 ventricle, respectively), the functional benefit of TLV was combined to a potent reduction in
189 infarct size vs Saline and Control groups (Figure 5A). Myocardial necrosis out of the risk

190 zone was also significantly limited by TLV as compared to Control and Saline groups (Figure
191 5, Panels B-D).

192 TLV limits lung cardiogenic lesions

193 As shown in Table IV, a metabolic acidosis was observed in all groups after cardiac
194 arrest. One hour after coronary reperfusion, blood PO₂ and pH were improved in TLV vs
195 Control and Saline groups. At necropsy, lung examination showed reduced lung cardiogenic
196 lesions in TLV vs Control and Saline (Figure 6, Panels A-C). Foam macrophages were
197 identified in the TLV group in 8/10 animals (Figure 6, Panel D), probably as a consequence
198 of some perfluorocarbons persistence. Kidney and liver dysfunctions and lesions were
199 moderate in all groups (Table II and Figure 6, Panels E-H).

200 TLV limits neurological dysfunction

201 As illustrated in Figure 7 (Panel A), the neurological deficit score was significantly
202 attenuated in the TLV group as compared to Control and Saline groups. This neuroprotective
203 effect was also supported by limited brain ischemic lesions in TLV vs Control and Saline
204 groups (Figure 7, Panels B-C). Large cerebral infarctions were observed in two rabbits from
205 the Control and Saline groups, as well as one small and deep infarction in the TLV group.

206 **Discussion**

207 In the present study, we demonstrate that hypothermic TLV not only attenuates
208 neurological injury and myocardial infarct size but also prevents acute cardiovascular
209 dysfunction and mortality after combined CAO and cardiac arrest. The favourable effect of
210 TLV was related to cooling rapidity as a slower cooling was not similarly efficient, despite
211 similar target temperature (32°C).

212 In this study, we observed a severe post-cardiac arrest syndrom (1) with an intense
213 cardiovascular dysfunction as acute CAO and cardiac arrest were associated. In Control
214 conditions, mortality within the first 24 h reached 60% while it was only 30% in a previous
215 report in a similar rabbit model with 10 min of shockable cardiac arrest without CAO (12).
216 This model is especially relevant for evaluation of acute heart failure and myocardial stunning
217 (15) after cardiac arrest. During the early phase of global stunning (*i.e.* prior to coronary
218 reperfusion), a favourable effect of ultra-fast cooling was evidenced with TLV. This was
219 probably a direct effect of hypothermia on ventricular contractility (16, 17) as previously
220 observed during regional stunning in rabbits submitted to focal myocardial ischemia (5). In
221 the present study, TLV further allowed an ultimate complete recovery of left ventricular
222 function within the first 24 h, as probably related to its potent anti-infarct effect. These
223 functional benefits were largely related to the rapidity of cooling since conventional and
224 slower hypothermia did not provide similar effects on cardiac output and cardiovascular
225 mortality. These results are in line with those of Yannopoulos et al. comparing cold saline
226 infusion to endovascular cooling in a swine model of ischemic cardiac arrest (18). Infarct size
227 reduction was again directly related to the time to reach the target temperature. In
228 comparison, our present study further demonstrates that the rapidity of cooling is not only
229 critical for infarct size reduction but more importantly for prevention of cardiovascular
230 mortality. The hemodynamic improvement afforded by TLV was however poorly predicted by
231 blood lactate levels.

232 Beyond its beneficial effect on cardiovascular outcomes, this study also evidenced
233 the neuroprotective effect of TLV through enhanced neurological recovery and decreased

234 brain lesions. Conversely, conventional cooling was not as protective, as previously shown in
235 a pure model of ventricular fibrillation (12). A benefit should however be evidenced with a
236 larger number of animals as even “slow” hypothermia is known to afford a neurological
237 benefit after cardiac arrest, although attenuated as compared to early hypothermia (7-10).
238 This clearly contrasts with the cardioprotective effect of hypothermia that is virtually entirely
239 lost when cooling is delayed (4-6). As an example, Knafelj et al. did not show any
240 cardioprotective benefit of slow and externally-induced hypothermia in survivors of ventricular
241 fibrillation with ST-elevation acute myocardial infarction (STEMI), whereas cerebral
242 performance category score was significantly improved (19). Clinical trials testing
243 hypothermia in STEMI patients with no cardiac arrest also showed disappointing results when
244 hypothermia was delayed (20) as compared to more precocious and aggressive cooling (21).

245 Taken together, all these findings clearly emphasized the major impact of the rapidity
246 of cooling on outcome after combined CAO and cardiac arrest. As also observed in pigs (22,
247 23) and despite its invasive nature, hypothermic TLV seems to provide additional benefits to
248 postresuscitation care. In the latter studies, some lung toxicity was however observed with
249 the perfluorocarbon Fluorinert™ FC-77. The current development of liquid ventilators
250 appropriate for human uses will allow confirmation of the present results for a further clinical
251 translation (24). In the present study, we used perfluorodecalin for TLV as this compound
252 was recently approved for intrapulmonary administration and bronchoalveolar lavage
253 (Perflubronc PFD®, Origen, Austin, Texas, USA). Perfluorodecalin is also radiolucent while
254 the well-known perfluorooctylbromide (Perflubronc PFB®) is opaque on X ray with potential
255 disadvantages in case of a coronary angioplasty is required in future translations.
256 Perfluorodecalin is also a viscous liquid with a low vapour pressure as compared to other
257 perfluorocarbons. It will be necessary to test other perfluorocarbons with a higher vapour
258 pressure and evaporating rate (e.g., perfluorooctane). In the future, it will also be important to
259 confirm the present results in large animals as small ones poorly mimic human inflammation
260 states (25), that are known to be critical after cardiac arrest.

261 The present study has several limitations. First, a limited number of animals were
262 euthanized for ethical reasons before the end of the follow-up (i.e., 3 Control animals). This
263 was conducted after 48 h of follow-up when certain and rapid death was expected. Our
264 survival analyses therefore took into account spontaneous deaths but also decision to
265 sacrifice highly disabled animals in rare cases. Second, we did not measure coronary
266 perfusion pressure during the cardiac massage. It would be relevant to investigate the
267 massage efficiency.

268

269

270 **Conclusion**

271 In conclusion, ultra-fast cooling dramatically improved the cardiovascular outcome
272 after cardiac arrest with underlying coronary artery occlusion. Further reports should confirm
273 these results in larger species.

274

275

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347 mimic human inflammatory diseases. *Proc Natl Acad Sci U S A*. 2013; 110:3507-12.
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349

350 **Figures legends**

351

352 **Figure 1:** Schematic representation of the experimental protocol.

353 *CAO, coronary artery occlusion; CA, cardiac arrest; ROSC, Resumption of spontaneous*
354 *circulation; TLV, Total Liquid Ventilation; Saline, hypothermia induced by intravenous*
355 *administration of cold saline combined to external cooling*

356

357 **Figure 2:** Esophageal, tympanic and rectal temperatures.

358 *Statistical analyses was performed at baseline (B; n=10 in each group), at t=30 min of CAO*
359 *(n=10 in each group), at t=180 min of reperfusion (n=9, 10 and 9 in Control, TLV and Saline,*
360 *respectively) and t=480 min of reperfusion (n=7, 9 and 8 in Control, TLV and Saline,*
361 *respectively). See legend of Figure 1; *, p<0.05 vs corresponding Control; †, p<0.05 vs*
362 *corresponding Saline.*

363

364 **Figure 3:** Cardiac output, peripheral vascular resistances and left ventricular stroke volume.

365 *See legend of Figure 2.*

366 .

367 **Figure 4:**

368 ***Panel A:*** Kaplan-Meyer survival curves throughout the 7 days following coronary artery
369 occlusion and cardiac arrest in the different experimental groups.

370 ***Panel B:*** Number of animals deceased before 7 days of survival follow-up in each group,
371 according the suspected cause of death (cardiogenic vs neurological death).

372 *See legend of Figure 2.*

373

374

375 **Figure 5:**

376 **Panel A:** Infarct sizes (as percentage of the risk zone). Open and closed circles represent
377 individual and mean infarct sizes, respectively.

378 **Panel B:** Histopathological scores of the myocardium in a territory remote to the risk zone.
379 Lesions consisted of foci of necrosis (0=normal; 3 = frequent foci). Open circles represents
380 individual scores.

381 **Panel C:** Histopathological appearance of the myocardium remote to the risk zone in a rabbit
382 from the Control group.

383 **Panel D:** Normal histological appearance of the myocardium remote to the risk zone in a
384 rabbit from the TLV group. The blue dye stains microvessels out of the risk zone.

385 *See legend of Figure 2.*

386

387 **Figure 6:**

388 **Panels A and B:** Histopathological scores of cardiogenic lesions and infectious
389 complications in the lungs, respectively. Open circles represents individual scores.

390 **Panel C:** Histopathological appearance of the lung of a rabbit in the Control group. Lesions
391 consisted of extensive serous edema within the alveolae (arrows).

392 **Panel D:** Histopathological appearance of the lung of a rabbit in the TLV group. Foam
393 macrophages can be seen within the alveolae (arrows).

394 **Panels E and F:** Histopathological scores of alteration of the kidney and liver, respectively.
395 Open circles represents individual scores.

396 **Panel G:** Histopathological appearance of the kidney in the Control group. Lesions consisted
397 of mildly dilated proximal tubules.

398 **Panel H:** Histopathological appearance of the liver of a rabbit in the Control group. Lesions
399 consisted of systematized centrilobular congestion of the sinusoids.

400 *See legend of Figure 2.*

401

402 **Figure 7:**

403 **Panel A:** Neurological dysfunction scores at days 1 and 7 after coronary artery occlusion and
404 cardiac arrest in the different experimental groups. Open circles represent individual scores
405 and the thick line represents the median value of the corresponding group.

406 **Panel B:** Histopathological scores of alteration of the brain. Open circles represents
407 individual scores.

408 **Panel C:** Histopathological appearance of the brain in one rabbit of the Control group.
409 Lesions consisted in a cerebral infarction (arrow).

410 **Panel D:** Histopathological appearance of the hippocampus in one rabbit of the Saline group.

411 **Panel E:** Normal histological appearance of the hippocampus in one rabbit of the TLV group.

412 *See legend of Figure 2.*

413

Table I: Heart rate and mean arterial pressure

	Baseline	CAO		Coronary reperfusion			
		1 min	30 min	180 min	480 min	Day 1	Day 7
<i>Number of animals (n)</i>							
Control	10	10	10	9	8	4	0
TLV	10	10	10	10	9	9	8
Saline	10	10	10	9	9	6	3
<i>Heart rate (beats/min)</i>							
Control	253±25	241±24	205±21	224±21	256±25	263±67	-
TLV	257±25	234±30	145±19*†	161±21*	223±36	264±45	242±34
Saline	255±34	231±30	197±34	148±14*	242±30	255±50	241±47
<i>Mean arterial pressure (mmHg)</i>							
Control	78±11	60±16	89±12	79±9	78±8	70±15	-
TLV	80±13	60±12	82±10	83±4	74±8	72±6	68±14
Saline	77±9	55±11	81±5	79±5	77±9	77±12	74±14

TLV, total liquid ventilation; Saline, hypothermia induced by intravenous administration of cold saline combined to external cooling; *, $p < 0.05$ vs corresponding Control; †, $p < 0.05$ vs corresponding Saline.

Table II: Troponin I, creatinine and alanine-aminotransferase (ALAT) blood levels

	Baseline	Coronary reperfusion		
		60 min	360 min	Day 1
<u>Number of animals (n)</u>				
Control	9	9	8	4
TLV	10	10	9	9
Saline	10	9	8	7
<u>Troponine I blood levels (ng/ml)</u>				
Control	< 0.2	151±62	147±58	111±50
TLV	< 0.2	8±11 *†	75±83	28±40 *†
Saline	< 0.2	61±27 *	152±62	91±46
<u>Creatinine plasma levels (mg/l)</u>				
Control	5.9±1.3	9.3±2.2	8.5±2.9	9.4±5.2
TLV	6.8±1.6	7.8±2.8	7.3±1.7	8.5±2.8
Saline	7.4±2.7	6.4±3.5	5.2±2.9	6.1±3.2
<u>Alanine-aminotransferase plasma levels (U/l)</u>				
Control	26±13	74±36	79±48	79±28
TLV	33±20	59±39	87±64	83±78
Saline	28±9	61±33	83±58	61±29

See legend of Table I.

Table III: Hemodynamic parameters one week after cardiac arrest in surviving animals.

In order to determine “usual” values, parameters were also assessed in Sham rabbits not submitted to cardiac arrest.

	Control	TLV	Saline	Sham
Number of animals (n)	0	8	2	8
Left ventricular maximal rate of pressure development (dP/dt _{max})	-	4908±1130	1848±815	5535±2318
Left ventricular minimal rate of pressure development (dP/dt _{min})	-	4270±1166	-1317±499	5339±1376
Left ventricular maximal systolic pressure (mmHg)	-	102±20	60±4	97±17
End-diastolic left ventricular pressure (mmHg)	-	6±2	14±6	4±1

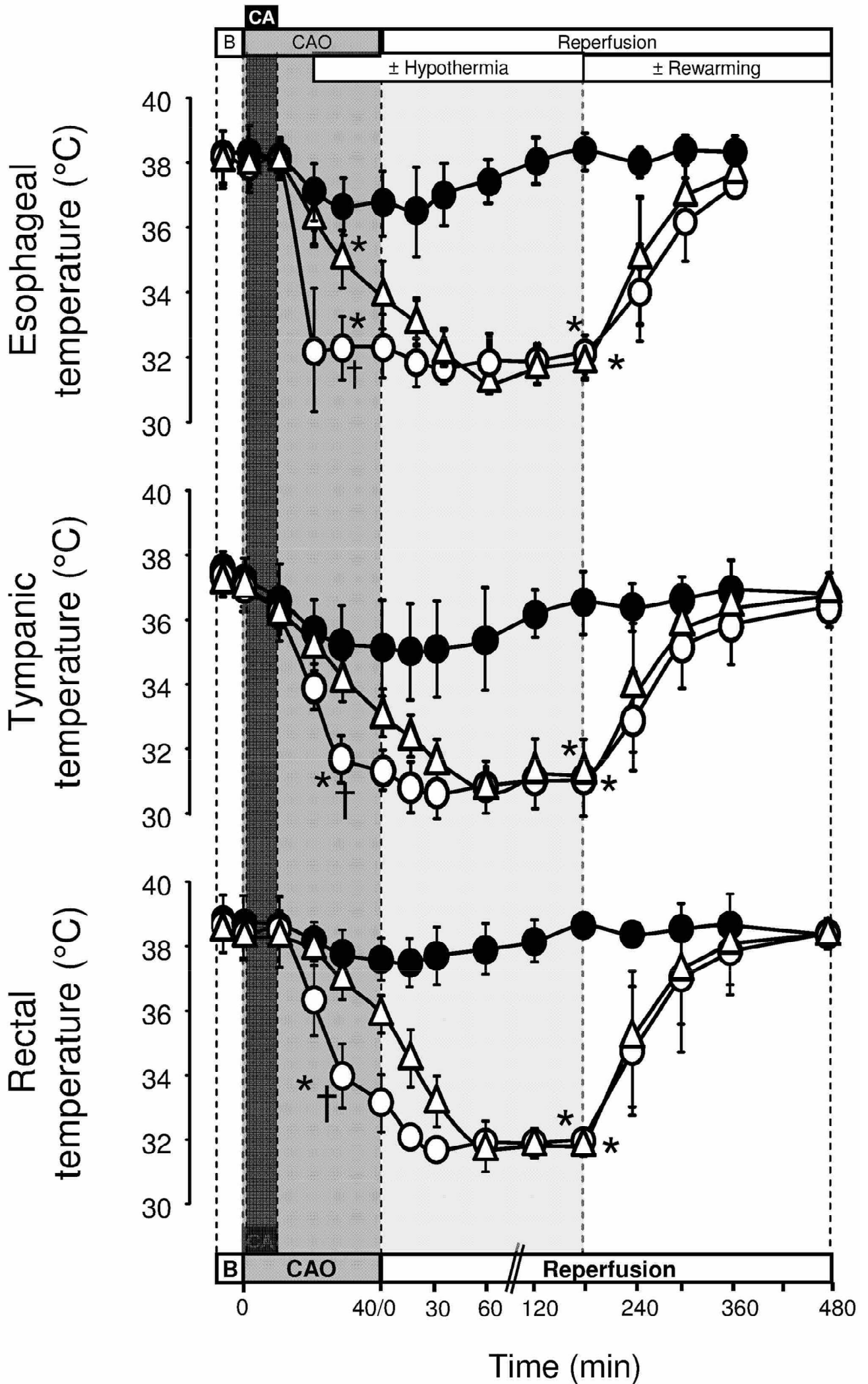
See legend of Table I. Statistical comparisons were only performed between TLV and Sham groups (no difference); One rabbit died in the Saline group during anaesthesia before catheterization.

Table IV: Blood gases and lactate blood levels.

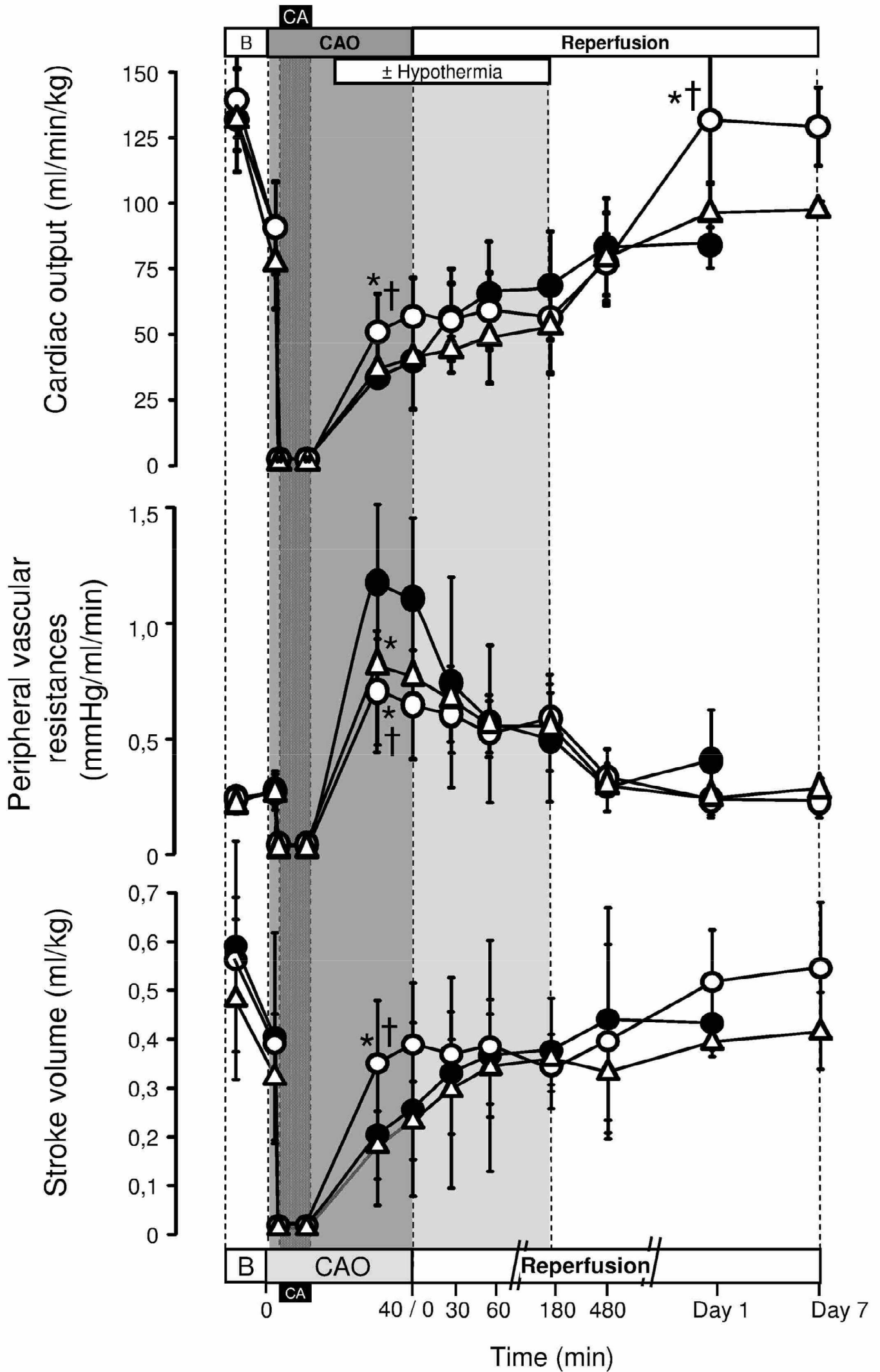
	Baseline	Coronary reperfusion			
		60 min	180 min	Day 1	Day 7
<i><u>Number of animals (n)</u></i>					
Control	10	10	9	4	0
TLV	10	10	10	9	8
Saline	10	10	9	6	3
<i><u>Lactates blood levels (mmol/l)</u></i>					
Control	3.0±0.9	13.5±3.1	11.0±3.2	3.3±2.9	-
TLV	3.4±0.8	13.7±3.3	12.8±3.5	2.3±1.1	3.7±1.2
Saline	3.5±1.6	16.1±2.1	15.7±2.9 *	4.5±4.2	4.7±3.2
<i><u>Blood pH</u></i>					
Control	7.41±0.07	7.03±0.23	7.22±0.1	7.31±0.07	-
TLV	7.45±0.11	7.33±0.09 *†	7.29±0.15	7.32±0.15	7.38±0.06
Saline	7.38±0.06	7.15±0.09	7.15±0.09	7.21±0.11	7.36±0.09
<i><u>Blood pCO₂ (torr)</u></i>					
Control	40±8	41±7	38±4	57±2	-
TLV	42±10	28±5 *	26±4 *	53±10	43±5
Saline	42±13	27±5 *	25±4 *	57±10	52±10
<i><u>Blood pO₂ (torr)</u></i>					
Control	428±129	205±95	373±136	57±9	-
TLV	500±170	456±126 *†	354±204	56±18	78±19
Saline	526±76	283±121	305±114	46±10	67±26

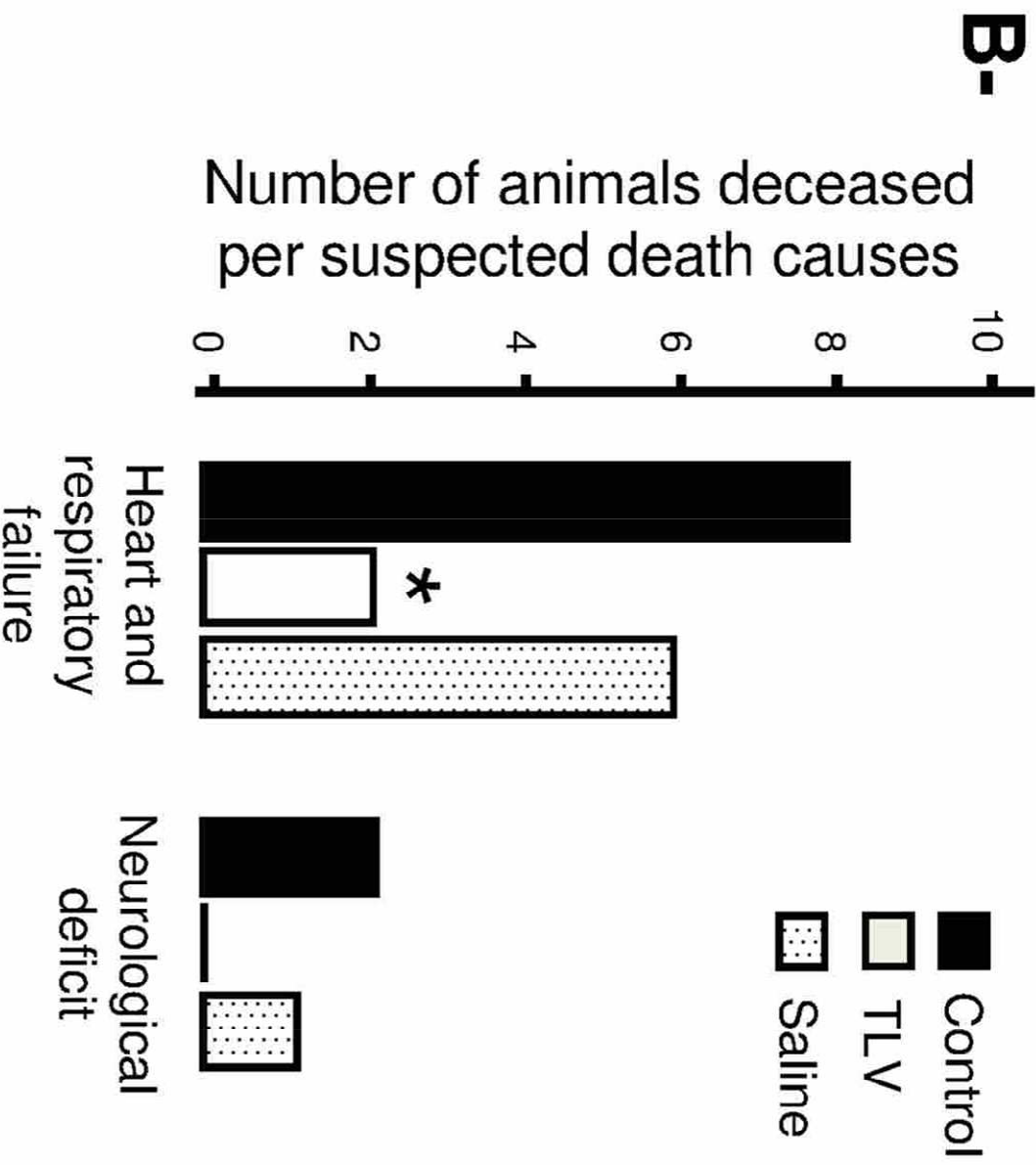
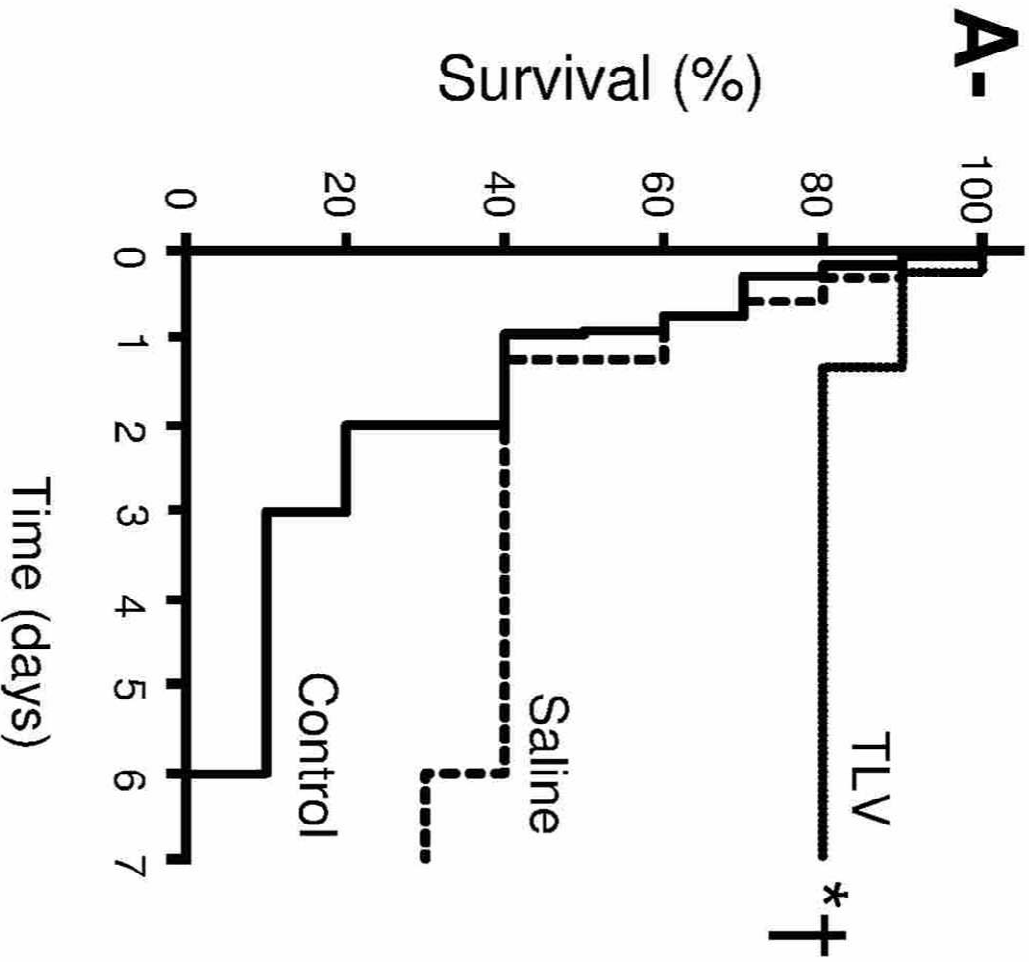
See legend of Table I.

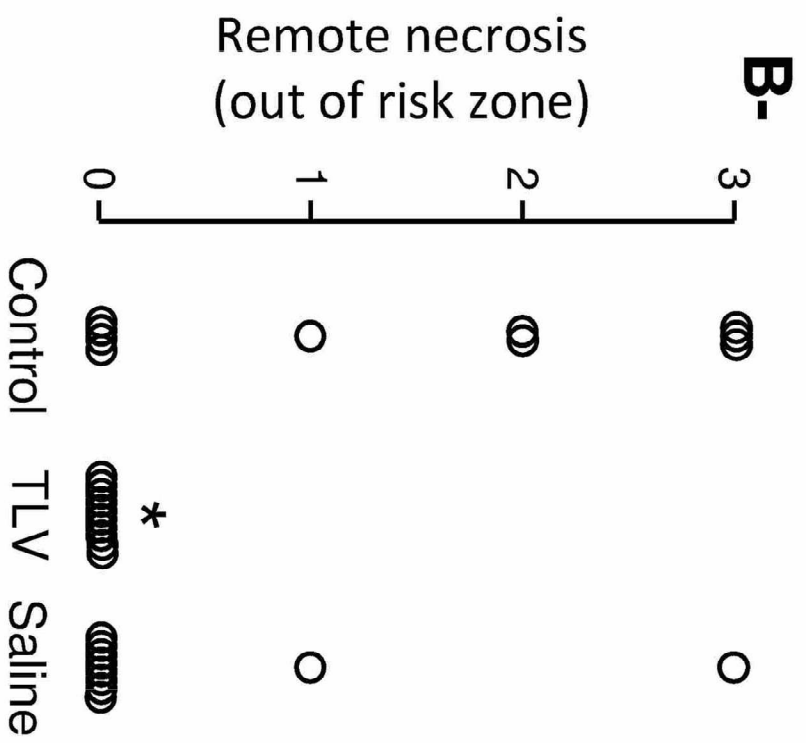
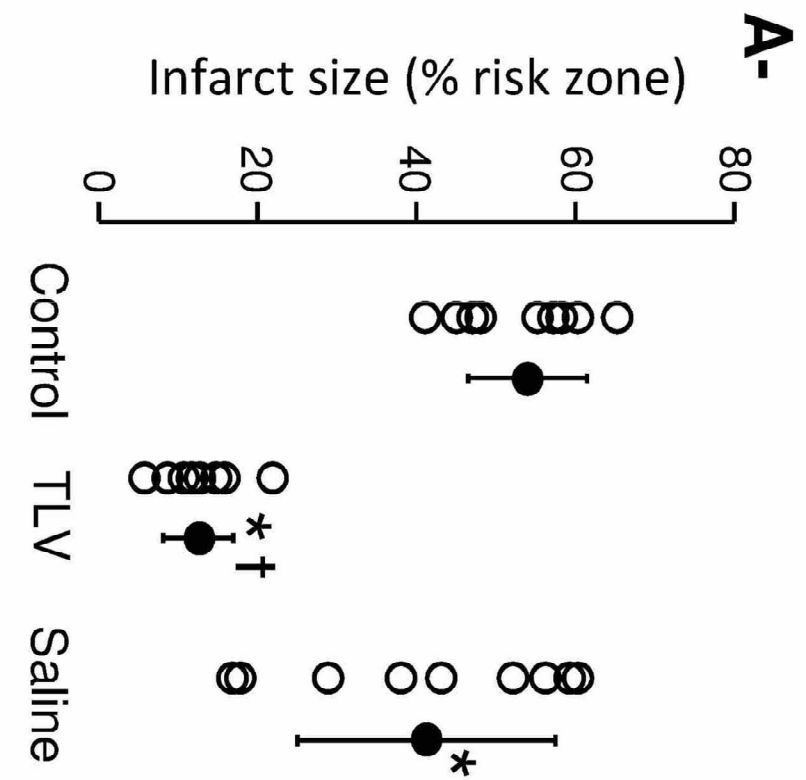
● Control ○ TLV △ Saline



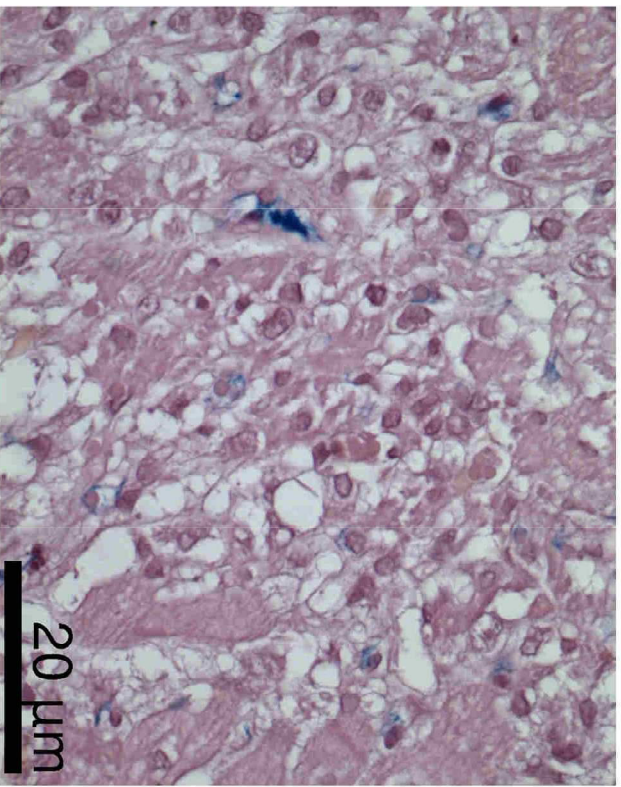
● Control ○ TLV ▲ Saline







C-



D-

