



**HAL**  
open science

## The small chill: mild hypothermia for cardioprotection?

Renaud Tissier, Mourad Chenoune, Bijan Ghaleh, Michael V. Cohen, James M. Downey, Alain Berdeaux

### ► To cite this version:

Renaud Tissier, Mourad Chenoune, Bijan Ghaleh, Michael V. Cohen, James M. Downey, et al.. The small chill: mild hypothermia for cardioprotection?: Cooling the ischemic myocardium. *Cardiovascular Research*, 2010, 88 (3), pp.406-14. 10.1093/cvr/cvq227 . inserm-00507838

**HAL Id: inserm-00507838**

**<https://www.hal.inserm.fr/inserm-00507838>**

Submitted on 11 Jul 2011

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

## **The Small Chill: mild hypothermia for cardioprotection?**

Renaud TISSIER<sup>1,2,3</sup>, Mourad CHENOUNE<sup>1,2,3</sup>, Bijan GHALEH<sup>1,2,3</sup>,  
Michael V COHEN<sup>4,5</sup>, James M DOWNEY<sup>4</sup>, Alain BERDEAUX<sup>1,2,3</sup>

1- INSERM, Unité 955, Equipe 3, Créteil, F-94000 France

2- Ecole Nationale Vétérinaire d'Alfort, Maisons-Alfort, F-94704 France

3- Université Paris Est, Créteil, F-94000 France

4- Department of Physiology, University of South Alabama, College of Medicine, Mobile,  
AL 36688 USA

5- Department of Medicine, University of South Alabama, College of Medicine, Mobile, AL  
36688 USA

**Running title: Cooling the ischemic myocardium**

**Word counts: 7521**

**Corresponding author:**

Dr Renaud Tissier

INSERM U955, équipe 3

Faculté de Médecine, Université Paris Est Créteil

8 rue du Général Sarrail

94010 Créteil cedex, France

Tel: +33.1.49.81.36.51 ; Fax: +33.1.48.99.17.77

E-mail: [rtissier@vet-alfort.fr](mailto:rtissier@vet-alfort.fr)

**Key words**

Cardioprotection

Cooling

Hypothermia

Infarction

Ischemia

**Conflict of Interest: none declared**

## 1 **1- Introduction**

2 Therapeutic whole body hypothermia has been considered for centuries.<sup>1</sup> For  
3 example, the Russian approach to resuscitation of a patient in cardiac arrest in 1803  
4 consisted of covering him with snow and then hoping for the resumption of a spontaneous  
5 circulation.<sup>1</sup> A century and a half later, the beneficial effect of therapeutic hypothermia  
6 during cardiac surgery was proven in canine models.<sup>2,3</sup> Outside of the operating room,  
7 hypothermia has been demonstrated to protect the brain following cardiac arrest in both  
8 animal models and humans.<sup>4,5</sup> The American Heart Association and the European  
9 Resuscitation Council both recommend the use of hypothermia in comatose patients  
10 resuscitated from cardiac arrest to improve the subsequent neurological recovery.<sup>6,7</sup>

11 Beside hypothermia's beneficial effect during cardiac surgery or following cardiac  
12 arrest, it has also been clearly demonstrated that even very mild hypothermia can greatly  
13 increase the heart's tolerance to myocardial ischemia resulting in decreases of infarct size in  
14 animal models of coronary artery occlusion. Mild hypothermia (body temperature down to  
15 32 °C) has the advantage that the heart continues to pump normally and no extracorporeal  
16 support is needed. However, despite the encouraging results in animals, clinical trials of  
17 mild hypothermia in patients being revascularized for acute myocardial infarction have  
18 yielded surprisingly disappointing effects on infarct size.<sup>8-10</sup> One goal of the present review  
19 is to consider why past trials have failed and propose what might be done to make cooling-  
20 induced cardioprotection more effective. We will review the literature to see if an optimal  
21 target temperature and cooling method can be recommended. Finally, the mechanism of  
22 mild cooling's protection will be revisited. While deep hypothermia stops the heart and  
23 clearly preserves ATP during ischemia, cooling to 34°C has only a modest effect on ATP  
24 depletion during ischemia<sup>11</sup> but it is as protective to the rabbit heart as is ischemic  
25 preconditioning.<sup>12</sup> That has caused some to suspect that the mechanism of the

26 cardioprotective effect of mild hypothermia might be more complex than energy  
27 preservation alone.<sup>13</sup>

28

## 29 **2. Physiological effects of mild hypothermia**

30 Hypothermia has distinct physiological effects. Hypothermia can be classified as  
31 mild (32-35°C), moderate (28-32°C), severe (20-28°C) or profound (<20°C). Mild  
32 hypothermia is the only one to be used for a whole-body therapeutic purpose as lower  
33 temperatures are increasingly life-threatening. Below 28°C, the heart spontaneously  
34 fibrillates in most mammalian species. Conversely, mild whole-body hypothermia is  
35 remarkably well tolerated by both animals and humans. The physiological effects of mild  
36 hypothermia include a decrease in heart rate and a subsequent fall in cardiac output (-7% for  
37 each °C),<sup>3, 14</sup> while stroke volume and mean arterial pressure remain unchanged. Cooling of  
38 the skin provokes an increase in systemic vascular resistance. With respect to regional  
39 hemodynamics, cerebral blood flow and intracranial pressure decrease,<sup>3, 14</sup> whereas renal  
40 blood flow and subsequent diuresis are increased.<sup>15</sup> Mild hypothermia also attenuates the  
41 cerebral metabolic rate (-6 to -7% for each °C).<sup>16</sup> Intestinal motility is also depressed by  
42 mild hypothermia. Blood pH changes by +0.016 unit for each decrease of 1°C, and serum  
43 potassium decreases as a result of its enhanced cellular uptake.<sup>17</sup> Hypothermia-induced  
44 hypokalemia should be corrected with caution during hypothermia since raising serum K<sup>+</sup>  
45 usually leads to hyperkalemia during subsequent rewarming. Other adverse effects include  
46 increased infection risk, a mild coagulopathy<sup>17</sup> and hyperglycemia caused by decreased  
47 plasma insulin levels.<sup>18</sup>

48

## 49 **3. The infarct limiting property of cooling: experimental *in vivo* investigations**

50 *3.1. Proof of concept of the myocardial infarct-limiting property of hypothermia*

51 Heart temperature is known to be a major determinant of infarct size in animal  
52 models of acute myocardial ischemia.<sup>19-21</sup> As an example, infarct size resulting from 30 min  
53 of regional myocardial ischemia in *in situ* rabbit hearts decreases by 8% of the risk zone for  
54 each °C decrement.<sup>21</sup> The infarct-limiting property of mild hypothermia has been confirmed  
55 in several species including dogs,<sup>19</sup> rabbits,<sup>12, 21-25</sup> pigs<sup>20, 26-28</sup> and rats.<sup>29</sup>

### 56 3.2. *What is the optimal target temperature?*

57 Importantly, there is no threshold temperature for the cardioprotective effect of  
58 hypothermia.<sup>19-21</sup> In fact, warming the heart above normothermia extends infarct size by the  
59 same degree as cooling reduces it.<sup>21</sup> For example, Hamamoto et al.<sup>30</sup> demonstrated in sheep  
60 submitted to 60 min of coronary artery occlusion that infarct size was progressively reduced  
61 when cardiac temperature during ischemia and reperfusion was lowered by 1°C decrements  
62 from 39.5 to 35.5°C. As shown in Table I, temperature decrement during ischemia within  
63 the mild hypothermic range has demonstrated a powerful anti-infarct effect in many studies.  
64 Figure 1 illustrates cooling to only 35°C at the onset of ischemia elicits ~70% decrease in  
65 infarct size following 30 min of regional ischemia in the open-chest rabbit.<sup>12</sup> And protection  
66 is still realized if the onset of cooling is delayed for 10 min. Cooling the heart to 32°C at the  
67 onset of ischemia essentially prevented infarction, and a protective effect was still evident  
68 when cooling was initiated as long as 20 min after the onset of ischemia and, therefore, 10  
69 min before reperfusion.<sup>12</sup> Furthermore, even when the ischemic period was extended to 60-  
70 120 min and cooling to 30-32°C was initiated 20-30 min after the onset of ischemia, the  
71 infarction process appeared to be halted when cooling was started and significant  
72 myocardial salvage was realized.<sup>12, 22</sup> And the heart continued to beat strongly and support  
73 the rabbit's circulation.

### 74 3.3. *What is the window of protection?*

75 Cooling the heart during the ischemic period reduces infarct size. Very early studies  
76 from the Corday laboratory<sup>31, 32</sup> clearly showed that if the dog heart were retrogradely

77 perfused with cooled arterial blood through the coronary sinus beginning 30 min after  
78 coronary occlusion and persisting for the remaining 2½-3 hr of the ischemic period, infarct  
79 size was decreased by 65-90%. Table 1 reveals that the degree of protection afforded by  
80 cooling is not only determined by the depth of cooling but also by its duration during the  
81 ischemic period. Miki et al.<sup>12</sup> demonstrated, for example, that the magnitude of infarct size  
82 reduction decreased when the onset of hypothermia was delayed from the beginning to the  
83 20<sup>th</sup> min of a 30-min period of ischemia in rabbits. As illustrated in Figure 1, cooling during  
84 just the last 10 minutes of ischemia still afforded significant cardioprotection when the target  
85 temperature was 32°C but not when it was 35°C. Figure 2 merges the results of several  
86 studies investigating the effect of hypothermia to 32°C initiated at different times during a  
87 30-min period of ischemia in rabbits, again illustrating that cardioprotection is attenuated  
88 when the onset of cooling is delayed. This raises a major logistical problem in the clinical  
89 setting since these patients with acute myocardial infarction present with ischemia already in  
90 progress. To significantly shorten the normothermic ischemic time cooling would have to  
91 be accomplished very soon after arrival in the hospital and efforts to effect early cooling  
92 must not come at the expense of increasing the time to reperfusion. That schedule has been  
93 difficult to implement.

94 Cooling the body at the time of reperfusion or beyond seems to be protective in the  
95 central nervous system.<sup>3,33</sup> That has led investigators to ask whether mild hypothermia  
96 might be cardioprotective when instituted only at the onset of the reperfusion phase, i.e., a  
97 postconditioning maneuver. Cooling to 10°C at the onset of reoxygenation and glucose  
98 resupply of chick cardiomyocytes subjected to simulated ischemia and reperfusion improved  
99 cell viability, and initiation of cooling towards the end of the simulated ischemia further  
100 diminished cell necrosis.<sup>34</sup> These observations strongly suggested that cooling during the  
101 peri-reperfusion period might be successful as well. Table 2 shows the results of several  
102 intact animal studies that have investigated this possibility.<sup>25, 28, 35-39</sup> Unfortunately, all but

103 one<sup>37</sup> failed to see any effect on infarct size and in the one positive study cooling still  
104 included that last 5 min of a 30 min ischemic period. Either the much lower temperature or  
105 the non-mammalian cell type was likely responsible for the protection in the chick  
106 cardiomyocyte study.<sup>34</sup> Of course, cooling a patient to 10°C is not feasible because that  
107 would stop the heart. However, it was found that hypothermia during reperfusion prevents  
108 microvascular damage and thereby offers some protection against the no-reflow  
109 phenomenon.<sup>24, 35</sup> One might, therefore, speculate that hypothermia at reperfusion protects  
110 against vascular alteration and subsequent microvascular obstruction<sup>35</sup> but cannot protect the  
111 cardiomyocytes. This could mean that one should maintain cooling during the first hours of  
112 reperfusion even if cooling had been instituted early in the ischemic period. Surprisingly,  
113 the question of whether prolonging hypothermia after reperfusion measurably adds to the  
114 anti-infarct effect has yet to be addressed. It should be noted that most patients spend  
115 approximately 90 minutes in hospital prior to revascularization because of delays associated  
116 with admitting, diagnosis, and preparation for intervention.<sup>40</sup> Whole-body cooling during  
117 that period could produce significant reduction of the resulting infarct.

#### 118 *3.4. How can myocardium be cooled?*

119 As emphasized earlier, the sooner hypothermia is achieved following the onset of  
120 ischemia, the more cardioprotective it would be. Experimentally, some studies have been  
121 performed using topical epicardial cooling to quickly lower cardiac temperature, but this  
122 method would be difficult to implement in patients. In patients with acute myocardial  
123 infarction, the critical parameters determining benefit would be the time when cooling could  
124 be started and also the rate at which cooling could be achieved. As explained earlier, the  
125 benefit would be expected to be small or absent if the target temperature is only reached far  
126 into the reperfusion phase. Obviously, the least invasive strategy for implementing  
127 therapeutic hypothermia in patients is external cooling. This can be done using ice packs or  
128 with specific medical devices designed to promote heat exchange by a more efficient



129 contact between the skin and the cooling medium.<sup>2, 41, 42</sup> Unfortunately the cooling rate  
130 afforded by these strategies is rather low, averaging only 1-2°C/h in humans.<sup>43</sup> That is  
131 because the cutaneous microcirculation tends to constrict when cold in an attempt to  
132 thermally insulate the body from the environment. Small laboratory animals cool faster with  
133 skin cooling than humans since their ratio of body mass to surface area is much smaller than  
134 in man. The time required to cool a rabbit weighing 2.0-3.0 kg to 32°C is still ~45 min, and  
135 this does not significantly limit infarct size after 60 min of ischemia.<sup>44</sup> While it is unlikely  
136 that patients with acute myocardial infarction would experience any anti-infarct benefit from  
137 conventional external cooling, this strategy is reported to be beneficial following cardiac  
138 arrest when even delayed hypothermia improves overall survival and neurological recovery  
139 after resuscitation.<sup>2, 41, 42</sup>

140 Other strategies have been proposed to induce therapeutic hypothermia. Some  
141 examples include endovascular cooling with intravenous thermodes, infusion of cold  
142 intravenous fluids, gastric lavage with cold fluid through a nasogastric catheter and even  
143 urinary bladder lavage.<sup>42</sup> Most of those techniques have been investigated in the clinics for  
144 their neuroprotective abilities.<sup>42</sup> The one that has been investigated for a cardioprotective  
145 therapy is endovascular cooling.<sup>26, 45, 46</sup> In human-sized pigs, this strategy afforded  
146 promising results when cooling was initiated early during ischemia.<sup>26, 45</sup> A clinical trial in  
147 patients with ST-segment elevation myocardial infarction (STEMI) clearly demonstrated the  
148 feasibility of that strategy,<sup>46</sup> but disappointingly did not show a significant cardioprotective  
149 benefit,<sup>8</sup> probably because of an insufficient cooling rate that resulted in normothermia  
150 during most of the ischemic period.<sup>47</sup>

### 151 *3.5. Ultra-fast cooling*

152 Accordingly other strategies have been proposed that elicit a much faster rate of  
153 cooling which should increase the degree of cardioprotection in the clinical setting.  
154 Examples include extracorporeal blood cooling<sup>12</sup> or total liquid ventilation with

155 temperature-controlled perfluorocarbons.<sup>25, 48</sup> These techniques can decrease cardiac  
156 temperature to 32°C within 3-5 min in rabbits. A pericardial perfusion circuit has also been  
157 proposed.<sup>49</sup> Unfortunately all these strategies would be challenging to implement in patients  
158 presenting with myocardial infarction since they are very invasive and would significantly  
159 delay reperfusion by angioplasty or thrombus extraction. A promising technique could be  
160 peritoneal lavage, which, although still invasive, should be easier to institute than any of the  
161 above-mentioned methods. As seen in patients with malignant hyperthermia it can cool a  
162 patient very quickly.<sup>50</sup> Whether any of these ultrafast cooling strategies would be beneficial  
163 in humans with STEMI remains to be investigated.

164

#### 165 **4- Mechanism of cooling-induced cardioprotection**

166 The mechanism of hypothermia-induced cardioprotection has mostly been  
167 investigated in experimental models receiving cold cardioplegia. The temperature of the  
168 heart is reduced to very low levels which, among other things, arrests it.<sup>13, 51</sup> However,  
169 mechanisms are likely to be quite different for mild hypothermia of an *in vivo* beating heart  
170 and an arrested heart exposed to cold cardioplegia. In beating hearts, one could, for example,  
171 argue that the cardioprotection might be related to the bradycardia elicited during ischemia.  
172 However, this is unlikely because the relationship between infarct size and temperature was  
173 unchanged when normothermic heart rate was restored by pacing in hypothermic rabbits.<sup>21,</sup>  
174 <sup>52</sup> It is therefore commonly assumed that hypothermia protects the heart, at least in part,  
175 through reduced cardiac metabolism. This assumption has been amply supported by studies  
176 performed during cold cardioplegia (<20°C).<sup>51, 53-60</sup> Most enzymes have a Q<sub>10</sub> of about 2,  
177 which means the reaction rate doubles for every 10°C increase in temperature. Reducing the  
178 cardiac temperature by 20°C should therefore decrease metabolism by a factor of 4. Mild  
179 hypothermia (>30°C) also decreases the rate of high energy phosphate<sup>11, 61, 62</sup> and glucose<sup>63</sup>  
180 utilization as well as lactate accumulation<sup>63</sup> but to a lesser extent than deep hypothermia.

181           Deep hypothermia may also alter ion exchange because it inhibits  $\text{Na}^+/\text{Ca}^{2+}$  and  
182  $\text{Na}^+/\text{K}^+$  sarcoplasmic exchangers, although it paradoxically activates the  $\text{Na}^+/\text{H}^+$   
183 exchanger.<sup>64</sup> Interestingly, hibernating hypothermic frogs increase their resistance to  
184 hypoxia through a decreased demand for ATP by reduced  $\text{Na}^+/\text{K}^+$ -ATPase activity.<sup>65</sup> An  
185 NMR study in isolated newborn rabbit hearts further confirmed that deep cooling (12°C)  
186 with cold crystalloid cardioplegia limited acidosis and calcium and sodium cellular overload  
187 during ischemia/reperfusion.<sup>13</sup> Deep hypothermia (17°C) also limited mitochondrial calcium  
188 overload in Langendorff guinea pig hearts undergoing ischemia.<sup>66</sup> While deep hypothermia  
189 tends to increase baseline reactive oxygen species formation during normoxia, it limits the  
190 burst following ischemia-reperfusion.<sup>66</sup> Using electron spin resonance spectroscopy in  
191 isolated reperfused rat heart investigators have observed reduced free-radical generation at  
192 reperfusion following ischemia at 4°C.<sup>67</sup>

193           Deep hypothermia reduces several modulators of the mitochondrial permeability  
194 transition pore (mPTP) *i.e.*, ATP depletion, calcium overload, and generation of reactive  
195 oxygen species. Mild hypothermia (32°C) inhibited calcium-induced mPTP opening in  
196 ventricular samples from rabbit hearts subjected to ischemia alone or to ischemia followed  
197 by 10 min of reperfusion.<sup>48</sup> Suppression of mPTP formation at reperfusion is thought to be  
198 the mode of action of ischemic preconditioning<sup>68</sup> and ischemic postconditioning.<sup>69</sup> Thus it  
199 is reasonable to speculate that hypothermia acts, at least in part, through inhibition of MPTP  
200 formation. However, the manner of modulation of mPTP opening is probably different than  
201 that afforded by pre- and postconditioning since the latter strategies are believed to trigger  
202 signal transduction pathways that determine the fate of the previously ischemic myocardium  
203 during the first minutes of reperfusion,<sup>69-73</sup> while hypothermia seems to exert its protection  
204 during ischemia.<sup>48</sup> Opening of mPTP at reperfusion only occurs if the heart has been injured  
205 by a period of prolonged ischemia. The nature of that injury is poorly understood, but Honda

206 and colleagues<sup>74</sup> referred to it as “priming”. Mild hypothermia may act to lessen that  
207 ischemic injury.

208         The protection from mild hypothermia is proportional to the decrease in temperature  
209 which argues against any off/on type of mechanism. A direct effect of mild hypothermia on  
210 enzyme kinetics seems unlikely to be the protective mechanism since most enzymes are not  
211 so temperature-dependent. Hearts function quite well over the entire range of mild  
212 hypothermia (32-38°C). An intriguing possibility is that mild hypothermia might somehow  
213 activate cardioprotective signal transduction pathways. Ning et al.<sup>75</sup> demonstrated that cold  
214 cardioplegia (30°C) preserves mitochondrial protein gene expression during hypoxia,  
215 including genes coding for HSP70, ANT<sub>1</sub>, and β-F<sub>1</sub>-ATPase.<sup>76</sup> They observed that neither  
216 ATP levels nor anaerobic metabolism are linked to mRNA expression of these latter  
217 proteins.<sup>77</sup> Ning et al.<sup>77</sup> also showed that moderate hypothermia (30°C) promotes expression  
218 of proteins involved in cell survival, while it inhibits induction of p53 protein. It is,  
219 therefore, reasonable to hypothesize that mild hypothermia triggers its protection through  
220 thermal sensors. This hypothesis is supported by studies from Halestrap’s group which  
221 reported that a short period of perfusion at 26°C in isolated rat hearts induced a  
222 preconditioning-like protection which increased protein kinase C-ε translocation to the  
223 particulate fraction (an index of its activation) and phosphorylated AMP-activated protein  
224 kinase (AMPK).<sup>78</sup> In one recent study in chick cardiomyocytes undergoing simulated  
225 ischemia, cooling to 25°C just before reoxygenation protected the cells.<sup>34</sup> More importantly  
226 that protection could be blocked by either a PKC or a nitric oxide synthase (NOS) blocker  
227 indicating signal transduction. Protection in the latter study, however, seemed to mimic that  
228 of postconditioning rather than that of mild hypothermia since the critical time for cooling  
229 was during reoxygenation rather than simulated ischemia, and PKC and NOS are well  
230 known components of the signal transduction pathways of pre- and postconditioning. Also

231 cooling was much more severe. All chemical reactions are at some point temperature-  
232 dependent, but the temperature coefficients ( $Q_{10}$ ) for the various mammalian enzymes vary  
233 widely. The possibility that one particular enzyme might be very sensitive to temperature  
234 and could serve as a sensor to implement the cardioprotective effect of mild hypothermia  
235 through cell signaling is an attractive hypothesis, but of course that enzyme could be very  
236 difficult to find.

237 Hypothermia seems to protect against injury during ischemia, while ischemic  
238 preconditioning protects against injury during reperfusion. Because of the different  
239 mechanisms it is possible to add the two together to produce additive protection.<sup>12</sup> It is  
240 currently possible to pharmacologically postcondition a heart<sup>79, 80</sup> and such an intervention is  
241 thought to protect by a mechanism identical to that of preconditioning. Thus, it should be  
242 possible to combine mild hypothermia during ischemia with a postconditioning agent at  
243 reperfusion. The primary impediment to using mild hypothermia clinically has been the  
244 logistics of implementing it quickly after the onset of ischemia. However, if signaling  
245 pathways turn out to be responsible for mild hypothermia's protection, then pharmacological  
246 activation of those pathways would likely be simpler to implement and such an agent could  
247 possibly even be given by EMS personnel.

248

#### 249 **5- Cooling and cardioprotection: STEMI clinical data**

250 Soon after the turn of the 21st century the promising experimental results regarding  
251 the cardioprotective effect of cooling inspired several clinical trials testing mild  
252 hypothermia's feasibility and safety in STEMI patients.<sup>46, 81, 82</sup> As shown in Table 3, two  
253 large-scale clinical trials (COOL-MI<sup>8</sup> and ICE-IT<sup>10</sup>) were conducted using endovascular  
254 cooling. Both studies assessed infarct size using single photon emission computed  
255 tomography (SPECT). The results of these studies have not yet been published in peer-  
256 reviewed journals, but they have been summarized in several reviews.<sup>41, 83, 84</sup>

257 In the COOL-MI study (*COOLing as an adjunctive therapy to percutaneous*  
258 *intervention in patients with acute Myocardial Infarction*), 392 STEMI patients were  
259 enrolled within 6 hours following the onset of symptoms.<sup>8</sup> Patients were assigned to either  
260 treatment with a percutaneous coronary intervention alone or to cooling with an  
261 endovascular cooling device (Relieve Temperature Therapy System, Radiant Medical,  
262 Redwood City, CA, USA) prior to revascularization. The percutaneous coronary  
263 intervention was accomplished a mean of 18 min after cooling was instituted, with a mean  
264 temperature of 35.0°C at the moment of reperfusion. As shown in Table 3, cooling did not  
265 provide an overall significant reduction in infarct size, except in a sub-group of patients with  
266 anterior myocardial infarction that were cooled to less than 35°C at the time of  
267 revascularization.<sup>8</sup> The overall negative result of that study might have been biased since the  
268 average door-to-balloon time in cooled patients was 18 minutes longer than in control  
269 patients (110 vs 92 min).

270 In the ICE-IT study (Intravascular Cooling Adjunctive to Primary Coronary  
271 Intervention), the design was similar to COOL-MI with the inclusion of 228 patients  
272 randomized to either normothermic revascularization or to revascularization with mild  
273 hypothermia using another endovascular device (Innercool by Celsius Control System, San  
274 Diego, CA, USA).<sup>10</sup> Hypothermia again did not provide a significant benefit regarding  
275 infarct size in the whole population (Table 3). A trend for a benefit was observed in the sub-  
276 group with anterior infarction with a body temperature of less than 35°C at the time of  
277 reperfusion. Interestingly, a subanalysis of the 6 sites with the best protocol compliance out  
278 of the 22 participating sites also demonstrated a significant reduction in infarct size with  
279 cooling compared to conventional revascularization (-44%). These data suggest, as  
280 emphasized above, that the overall negative result of the COOL-MI and ICE-IT trials was  
281 related to a delay in institution of hypothermia and/or to an insufficient cooling rate which  
282 resulted in little shortening of the normothermic ischemic period. A further study (COOL-

283 MI II) was accordingly recently conducted with an earlier, deeper and faster cooling  
284 protocol, with cooling started in the emergency room rather than in the catheterization  
285 laboratory. The results of this last trial are not yet available.

286 It has conversely been suggested that hyperthermia may worsen the clinical outcome  
287 in patients with myocardial infarction.<sup>85</sup> In experimental conditions, Chien et al.<sup>21</sup> found not  
288 only that hypothermia could diminish infarct size but that hyperthermia could increase it.  
289 One would, therefore, speculate that therapeutic hypothermia would also be indicated to  
290 reverse the adverse effects of hyperthermia. A general relationship between clinical outcome  
291 and body temperature may well exist.

#### 292 *Cardiac Resuscitation*

293 Another setting in which the protective effect of cooling would be relevant is  
294 cardiopulmonary resuscitation. Cooling after the heart is restarted can indeed protect the  
295 central nervous system, as has been previously shown,<sup>5</sup> but it may also protect the heart since  
296 myocardial ischemia is a common cause of cardiac arrest.<sup>41, 84</sup> Theoretically, the cardiac  
297 benefit would even be greater for those patients than for “typical” STEMI as the time before  
298 revascularization is prolonged by the resuscitation time. There is controversy as to whether  
299 resuscitated patients should be immediately submitted to a coronary intervention<sup>41, 84</sup> since  
300 the use of hypothermia is basically recommended for comatose survivors after out-of-  
301 hospital cardiac arrest with ventricular fibrillation.<sup>6, 7</sup> Importantly, the combination of  
302 cooling and percutaneous intervention is at least feasible and should be safe in those  
303 patients.<sup>41, 84</sup>

304

#### 305 **6. Conclusion**

306 In conclusion, cooling the myocardium with whole body mild hypothermia is a very  
307 potent cardioprotective maneuver, at least in the experimental setting. The benefit depends  
308 upon the rapidity with which cooling is instituted and by how much it shortens the

309 normothermic ischemic time. To afford a clinical benefit, a cooling strategy should  
310 accordingly be designed to achieve the target temperature well before the time of  
311 revascularization. The depth of cooling is also important. Temperatures in the range of 32-  
312 35°C are considered safe. Yet these temperatures exert a powerful anti-infarct effect in  
313 animal studies. Finally, more studies of mild hypothermia are needed to determine its actual  
314 mechanism, as compared to deep hypothermia that acts through energy preservation.



315 **Funding**

316           This study was supported by grants HL-20648 and HL-50688 from the Heart, Lung  
317 and Blood Institute of the National Institutes of Health, grant TLVenCool (06-JCJC-0078)  
318 from the French “Agence Nationale pour la Recherche”, grant TLV-CARDAREST  
319 (R10028JS) from the INSERM-transfert and the ITMO “Technologies pour la Santé” and  
320 grant ET7-460 from the “Fondation de l’Avenir”. Mourad Chenoune was supported by a  
321 grant from the “Groupe de Reflexion sur la Recherche Cardiovasculaire” and by a “Poste  
322 d’accueil INSERM”.

323

324 **Acknowledgements**

325           We are greatly indebted to Fanny Lidouren and Alain Bizé for their excellent  
326 support.

## References

1. Liss HP. A history of resuscitation. *Ann Emerg Med* 1986;**15**:65-72.
2. Alzaga AG, Cerdan M, Varon J. Therapeutic hypothermia. *Resuscitation* 2006;**70**:369-380.
3. Varon J, Acosta P. Therapeutic hypothermia: past, present, and future. *Chest* 2008;**133**:1267-1274.
4. Bernard SA, Gray TW, Buist MD, Jones BM, Silvester W, Gutteridge G *et al.* Treatment of comatose survivors of out-of-hospital cardiac arrest with induced hypothermia. *N Engl J Med* 2002;**346**:557-563.
5. The Hypothermia After Cardiac Arrest Study Group. Mild therapeutic hypothermia to improve the neurologic outcome after cardiac arrest. *N Engl J Med* 2002;**346**:549-556.
6. Nolan JP, Morley PT, Vanden Hoek TL, Hickey RW, Kloeck WGJ, Billi J *et al.* Therapeutic hypothermia after cardiac arrest: an advisory statement by the advanced life support task force of the International Liaison Committee on Resuscitation. *Circulation* 2003;**108**:118-121.
7. Nolan JP, Deakin CD, Soar J, Böttiger BW, Smith G. European Resuscitation Council guidelines for resuscitation 2005. Section 4. Adult advanced life support. *Resuscitation* 2005;**67 (Suppl 1)**:S39-S86.
8. O'Neill WW on behalf of the COOL-MI Investigators. A prospective randomized trial of mild systemic hypothermia during PCI treatment of ST elevation myocardial infarction. *Presented at the 15<sup>th</sup> annual Transcatheter Cardiovascular Therapeutics, Washington, September 2003.* O'Neill WW, Dixon SR. The year in interventional cardiology. *J Am Coll Cardiol* 2004;**43**:875-890. 2003.
9. Stone GW, Dixon SR, Grines CL, Cox DA, Webb JG, Brodie BR *et al.* Predictors of infarct size after primary coronary angioplasty in acute myocardial infarction from pooled analysis from four contemporary trials. *Am J Cardiol* 2007;**100**:1370-1375.

10. Grines CL on behalf of the ICE-IT Investigators. Intravascular cooling adjunctive to percutaneous coronary intervention for acute myocardial infarction. *Presented at the 16<sup>th</sup> annual Transcatheter Cardiovascular Therapeutics, Washington DC, September 2004.*  
O'Neill WW, Dixon SR, Grines CL: The year in interventional cardiology. *J Am Coll Cardiol* 2005;45:1117-1134.
11. Jones RN, Reimer KA, Hill ML, Jennings RB. Effect of hypothermia on changes in high-energy phosphate production and utilization in total ischemia. *J Mol Cell Cardiol* 1982;14 (Suppl 3):123-130.
12. Miki T, Liu GS, Cohen MV, Downey JM. Mild hypothermia reduces infarct size in the beating rabbit heart: a practical intervention for acute myocardial infarction? *Basic Res Cardiol* 1998;93:372-383.
13. Anderson SE, Liu H, Beyschau A, Cala PM. Effects of cold cardioplegia on pH, Na, and Ca in newborn rabbit hearts. *Am J Physiol* 2006;290:H1090-H1097.
14. Bernard SA, Jones BMC, Horne MK. Clinical trial of induced hypothermia in comatose survivors of out-of-hospital cardiac arrest. *Ann Emerg Med* 1997;30:146-153.
15. Zeiner A, Sunder-Plassmann G, Sterz F, Holzer M, Losert H, Laggner AN *et al.* The effect of mild therapeutic hypothermia on renal function after cardiopulmonary resuscitation in men. *Resuscitation* 2004;60:253-261.
16. Rosomoff HL, Holaday DA. Cerebral blood flow and cerebral oxygen consumption during hypothermia. *Am J Physiol* 1954;179:85-88.
17. Polderman KH. Mechanisms of action, physiological effects, and complications of hypothermia. *Crit Care Med* 2009;37 (Suppl):S186-S202.
18. Lehot JJ, Piriz H, Villard J, Cohen R, Guidollet J. Glucose homeostasis. Comparison between hypothermic and normothermic cardiopulmonary bypass. *Chest* 1992;102:106-111.
19. Schwartz LM, Verbinski SG, Vander Heide RS, Reimer KA. Epicardial temperature is a major predictor of myocardial infarct size in dogs. *J Mol Cell Cardiol* 1997;29:1577-1583.

20. Duncker DJ, Klassen CL, Ishibashi Y, Herrlinger SH, Pavek TJ, Bache RJ. Effect of temperature on myocardial infarction in swine. *Am J Physiol* 1996;**270**:H1189-H1199.
21. Chien GL, Wolff RA, Davis RF, van Winkle DM. "Normothermic range" temperature affects myocardial infarct size. *Cardiovasc Res* 1994;**28**:1014-1017.
22. Hale SL, Kloner RA. Myocardial temperature reduction attenuates necrosis after prolonged ischemia in rabbits. *Cardiovasc Res* 1998;**40**:502-507.
23. Hale SL, Kloner RA. Myocardial hypothermia: a potential therapeutic technique for acute regional myocardial ischemia. *J Cardiovasc Electrophysiol* 1999;**10**:405-413.
24. Hale SL, Dae MW, Kloner RA. Hypothermia during reperfusion limits 'no-reflow' injury in a rabbit model of acute myocardial infarction. *Cardiovasc Res* 2003;**59**:715-722.
25. Tissier R, Hamanaka K, Kuno A, Parker JC, Cohen MV, Downey JM. Total liquid ventilation provides ultra-fast cardioprotective cooling. *J Am Coll Cardiol* 2007;**49**:601-605.
26. Dae MW, Gao DW, Sessler DI, Chair K, Stillson CA. Effect of endovascular cooling on myocardial temperature, infarct size, and cardiac output in human-sized pigs. *Am J Physiol* 2002;**282**:H1584-H1591.
27. Schwartz DS, Bremner RM, Baker CJ, Uppal KM, Barr ML, Cohen RG *et al.* Regional topical hypothermia of the beating heart: preservation of function and tissue. *Ann Thorac Surg* 2001;**72**:804-809.
28. Otake H, Shite J, Paredes OL, Shinke T, Yoshikawa R, Tanino Y *et al.* Catheter-based transcatheter myocardial hypothermia attenuates arrhythmia and myocardial necrosis in pigs with acute myocardial infarction. *J Am Coll Cardiol* 2007;**49**:250-260.
29. van den Doel MA, Gho BCG, Duval SY, Schoemaker RG, Duncker DJ, Verdouw PD. Hypothermia extends the cardioprotection by ischaemic preconditioning to coronary artery occlusions of longer duration. *Cardiovasc Res* 1998;**37**:76-81.

30. Hamamoto H, Sakamoto H, Leshnower BG, Parish LM, Kanemoto S, Hinmon R *et al.* Very mild hypothermia during ischemia and reperfusion improves postinfarction ventricular remodeling. *Ann Thorac Surg* 2009;**87**:172-177.
31. Haendchen RV, Corday E, Meerbaum S, Povzhitkov M, Rit J, Fishbein MC. Prevention of ischemic injury and early reperfusion derangements by hypothermic retroperfusion. *J Am Coll Cardiol* 1983;**1**:1067-1080.
32. Wakida Y, Haendchen RV, Kobayashi S, Nordlander R, Corday E. Percutaneous cooling of ischemic myocardium by hypothermic retroperfusion of autologous arterial blood: effects on regional myocardial temperature distribution and infarct size. *J Am Coll Cardiol* 1991;**18**:293-300.
33. Kollmar R, Schäbitz WR, Heiland S, Georgiadis D, Schellinger PD, Bardutzky J *et al.* Neuroprotective effect of delayed moderate hypothermia after focal cerebral ischemia: an MRI study. *Stroke* 2002;**33**:1899-1904.
34. Shao Z-H, Chang W-T, Chan KC, Wojcik KR, Hsu C-W, Li C-Q *et al.* Hypothermia-induced cardioprotection using extended ischemia and early reperfusion cooling. *Am J Physiol* 2007;**292**:H1995-H2003.
35. Götberg M, Olivecrona GK, Engblom H, Ugander M, van der Pals J, Heiberg E *et al.* Rapid short-duration hypothermia with cold saline and endovascular cooling before reperfusion reduces microvascular obstruction and myocardial infarct size. *BMC Cardiovasc Disord* 2008;**8**:7.
36. Hale SL, Dave RH, Kloner RA. Regional hypothermia reduces myocardial necrosis even when instituted after the onset of ischemia. *Basic Res Cardiol* 1997;**92**:351-357.
37. Kanemoto S, Matsubara M, Noma M, Leshnower BG, Parish LM, Jackson BM *et al.* Mild hypothermia to limit myocardial ischemia-reperfusion injury: importance of timing. *Ann Thorac Surg* 2009;**87**:157-163.

38. Maeng M, Mortensen UM, Kristensen J, Kristiansen SB, Andersen HR. Hypothermia during reperfusion does not reduce myocardial infarct size in pigs. *Basic Res Cardiol* 2006;**101**:61-68.
39. Schwiebert C, Huhn R, Heinen A, Weber NC, Hollmann MW, Schlack W *et al.* Postconditioning by xenon and hypothermia in the rat heart in vivo. *Eur J Anaesthesiol* 2010;**27**:000-000.
40. Magid DJ, Wang Y, Herrin J, McNamara RL, Bradley EH, Curtis JP *et al.* Relationship between time of day, day of week, timeliness of reperfusion, and in-hospital mortality for patients with acute ST-segment elevation myocardial infarction. *JAMA* 2005;**294**:803-812.
41. Holzer M, Behringer W. Therapeutic hypothermia after cardiac arrest and myocardial infarction. *Best Pract Res Clin Anaesthesiol* 2008;**22**:711-728.
42. Kimberger O, Kurz A. Thermoregulatory management for mild therapeutic hypothermia. *Best Pract Res Clin Anaesthesiol* 2008;**22**:729-744.
43. Hoedemaekers CW, Ezzahti M, Gerritsen A, van der Hoeven JG. Comparison of cooling methods to induce and maintain normo- and hypothermia in intensive care unit patients: a prospective intervention study. *Crit Care* 2007;**11**:R91.
44. Chenoune M, Lidouren F, Ghaleh B, Couvreur N, Dubois-Rande J-L, Berdeaux A *et al.* Rapid cooling of the heart with total liquid ventilation prevents transmural myocardial infarction following prolonged ischemia in rabbits. *Resuscitation* 2010;**81**:359-362.
45. Dae MW, Gao DW, Ursell PC, Stillson CA, Sessler DI. Safety and efficacy of endovascular cooling and rewarming for induction and reversal of hypothermia in human-sized pigs. *Stroke* 2003;**34**:734-738.
46. Dixon SR, Whitbourn RJ, Dae MW, Grube E, Sherman W, Schaer GL *et al.* Induction of mild systemic hypothermia with endovascular cooling during primary percutaneous coronary intervention for acute myocardial infarction. *J Am Coll Cardiol* 2002;**40**:1928-1934.

47. Wolfrum S, Radke PW, Schunkert H, Kurowski V. The authors reply to Ramaraj R: Mild therapeutic hypothermia in patients after out-of-hospital cardiac arrest due to acute ST-segment elevation myocardial infarction. *Crit Care Med* 2008;**36**:3280-3281.
48. Tissier R, Couvreur N, Ghaleh B, Bruneval P, Lidouren F, Morin D *et al.* Rapid cooling preserves the ischaemic myocardium against mitochondrial damage and left ventricular dysfunction. *Cardiovasc Res* 2009;**83**:345-353.
49. Dave RH, Hale SL, Kloner RA. Hypothermic, closed circuit pericardioperfusion: a potential cardioprotective technique in acute regional ischemia. *J Am Coll Cardiol* 1998;**31**:1667-1671.
50. Gjessing J, Barsa J, Tomlin PJ. A possible means of rapid cooling in the emergency treatment of malignant hyperpyrexia. *Br J Anaesth* 1976;**48**:469-473.
51. Stowe DF, Varadarajan SG, An J, Smart SC. Reduced cytosolic Ca<sup>2+</sup> loading and improved cardiac function after cardioplegic cold storage of guinea pig isolated hearts. *Circulation* 2000;**102**:1172-1177.
52. Hale SL, Kloner RA. Myocardial temperature in acute myocardial infarction: protection with mild regional hypothermia. *Am J Physiol Heart Circ Physiol* 1997;**273**:H220-H227.
53. van der Vusse GJ, van der Veen FH, Flameng W, Coumans W, Borgers M, Willems G *et al.* A biochemical and ultrastructural study on myocardial changes during aorto-coronary bypass surgery: St. Thomas Hospital cardioplegia versus intermittent aortic cross-clamping at 34 and 25 degrees C. *Eur Surg Res* 1986;**18**:1-11.
54. Smolenski RT, Lachno DR, Yacoub MH. Adenine nucleotide catabolism in human myocardium during heart and heart-lung transplantation. *Eur J Cardiothorac Surg* 1992;**6**:25-30.
55. Sukehiro S, Dyszkiewics W, Minten J, Wynants J, Van Belle H, Flameng W. Catabolism of high energy phosphates during long-term cold storage of donor hearts: effects



- of extra- and intracellular fluid-type cardioplegic solutions and calcium channel blockers. *J Heart Lung Transplant* 1991;**10**:387-393.
56. Bical O, Gerhardt M-F, Paumier D, Gaillard D, Comas J, Landais P *et al.* Comparison of different types of cardioplegia and reperfusion on myocardial metabolism and free radical activity. *Circulation* 1991;**84 (Suppl III)**:III-375-III-379.
57. Möllhoff T, Sukehiro S, Hendrickx M, Van Belle H, Flameng W. Effects of hypothermic ischemia on purine catabolism in canine, primate, and human myocardium. *Thorac Cardiovasc Surg* 1991;**39**:187-192.
58. Shirakura R, Matsuda H, Nakano S, Nakata S, Kaneko M, Takami H *et al.* Myocardial energy metabolism in asphyxiated canine hearts preserved for 24 hours. *Transplantation* 1992;**53**:1215-1218.
59. Chong YS, Cottier DS, Gavin JB. Myocardial protection during prolonged ischaemic cardiac arrest: experimental evaluation of three crystalloid cardioplegic solutions. *J Cardiovasc Surg* 1994;**35**:35-44.
60. Minten J, Flameng W, Dyszkiewicz W. Optimal storage temperature and benefit of hypothermic cardioplegic arrest for long-term preservation of donor hearts: a study in the dog. *Transpl Int* 1988;**1**:19-25.
61. Simkhovich BZ, Hale SL, Kloner RA. Metabolic mechanism by which mild regional hypothermia preserves ischemic tissue. *J Cardiovasc Pharmacol Ther* 2004;**9**:83-90.
62. Ning X-H, Xu C-S, Song YC, Xiao Y, Hu Y-J, Lupinetti FM *et al.* Hypothermia preserves function and signaling for mitochondrial biogenesis during subsequent ischemia. *Am J Physiol* 1998;**274**:H786-H793.
63. Ichihara K, Robishaw JD, Vary TC, Neely JR. Protection of ischemic myocardium from metabolic products. *Acta Med Scand Suppl* 1981;**651**:13-18.

64. Martineau Knerr SM, Lieberman M. Ion transport during hypothermia in cultured heart cells: implications for protection of the immature myocardium. *J Mol Cell Cardiol* 1993;**25**:277-288.
65. Boutilier RG. Mechanisms of cell survival in hypoxia and hypothermia. *J Exp Biol* 2001;**204**:3171-3181.
66. Riess ML, Camara AKS, Kevin LG, An J, Stowe DF. Reduced reactive O<sub>2</sub> species formation and preserved mitochondrial NADH and [Ca<sup>2+</sup>] levels during short-term 17 °C ischemia in intact hearts. *Cardiovasc Res* 2004;**61**:580-590.
67. Gambert S, Bès-Houtmann S, Vandroux D, Tissier C, Vergely-Vandriessse C, Rochette L *et al.* Deep hypothermia during ischemia improves functional recovery and reduces free-radical generation in isolated reperfused rat heart. *J Heart Lung Transplant* 2004;**23**:487-491.
68. Hausenloy DJ, Maddock HL, Baxter GF, Yellon DM. Inhibiting mitochondrial permeability transition pore opening: a new paradigm for myocardial preconditioning? *Cardiovasc Res* 2002;**55**:534-543.
69. Argaud L, Gateau-Roesch O, Raisky O, Loufouat J, Robert D, Ovize M. Postconditioning inhibits mitochondrial permeability transition. *Circulation* 2005;**111**:194-197.
70. Javadov SA, Clarke S, Das M, Griffiths EJ, Lim KHH, Halestrap AP. Ischaemic preconditioning inhibits opening of mitochondrial permeability transition pores in the reperfused rat heart. *J Physiol* 2003;**549**:513-524.
71. Gateau-Roesch O, Argaud L, Ovize M. Mitochondrial permeability transition pore and postconditioning. *Cardiovasc Res* 2006;**70**:264-273.
72. Hausenloy DJ, Duchon MR, Yellon DM. Inhibiting mitochondrial permeability transition pore opening at reperfusion protects against ischaemia-reperfusion injury. *Cardiovasc Res* 2003;**60**:617-625.

73. Cohen MV, Yang X-M, Downey JM. The pH hypothesis of postconditioning: staccato reperfusion reintroduces oxygen and perpetuates myocardial acidosis. *Circulation* 2007;**115**:1895-1903.
74. Honda HM, Korge P, Weiss JN. Mitochondria and ischemia/reperfusion injury. *Ann N Y Acad Sci* 2005;**1047**:248-258.
75. Ning X-H, Chen S-H, Xu C-S, Hyyti OM, Qian K, Krueger JJ *et al.* Hypothermia preserves myocardial function and mitochondrial protein gene expression during hypoxia. *Am J Physiol* 2003;**285**:H212-H219.
76. Ning X-H, Xu C-S, Portman MA. Mitochondrial protein and HSP70 signaling after ischemia in hypothermic-adapted hearts augmented with glucose. *Am J Physiol* 1999;**277**:R11-R17.
77. Ning X-H, Chi EY, Buroker NE, Chen S-H, Xu C-S, Tien Y-T *et al.* Moderate hypothermia (30°C) maintains myocardial integrity and modifies response of cell survival proteins after reperfusion. *Am J Physiol Heart Circ Physiol* 2007;**293**:H2119-2128.
78. Khaliulin I, Clarke SJ, Lin H, Parker J, Suleiman M-S, Halestrap AP. Temperature preconditioning of isolated rat hearts--a potent cardioprotective mechanism involving a reduction in oxidative stress and inhibition of the mitochondrial permeability transition pore. *J Physiol* 2007;**581**:1147-1161.
79. Gomez L, Thibault H, Gharib A, Dumont J-M, Vuagniaux G, Scalfaro P *et al.* Inhibition of mitochondrial permeability transition improves functional recovery and reduces mortality following acute myocardial infarction in mice. *Am J Physiol* 2007;**293**:H1654-H1661.
80. Piot C, Croisille P, Staat P, Thibault H, Rioufol G, Mewton N *et al.* Effect of cyclosporine on reperfusion injury in acute myocardial infarction. *N Engl J Med* 2008;**359**:473-481.

81. Kandzari DE, Chu A, Brodie BR, Stuckey TA, Hermiller JB, Vetrovec GW *et al.* Feasibility of endovascular cooling as an adjunct to primary percutaneous coronary intervention (results of the LOWTEMP pilot study). *Am J Cardiol* 2004;**93**:636-639.
82. Ly HQ, Denault A, Dupuis J, Vadeboncoeur A, Harel F, Arsenault A *et al.* A pilot study: the Noninvasive Surface Cooling Thermoregulatory System for Mild Hypothermia Induction in Acute Myocardial Infarction (the NICAMI Study). *Am Heart J* 2005;**150**:933.e9-933.e13.
83. Polderman KH. Induced hypothermia and fever control for prevention and treatment of neurological injuries. *Lancet* 2008;**371**:1955-1969.
84. Wolfrum S, Pierau C, Radke PW, Schunkert H, Kurowski V. Mild therapeutic hypothermia in patients after out-of-hospital cardiac arrest due to acute ST-segment elevation myocardial infarction undergoing immediate percutaneous coronary intervention. *Crit Care Med* 2008;**36**:1780-1786.
85. Naito K, Anzai T, Yoshikawa T, Maekawa Y, Sugano Y, Kohno T *et al.* Increased body temperature after reperfused acute myocardial infarction is associated with adverse left ventricular remodeling. *J Card Fail* 2007;**13**:25-33.

## **Legend of Figures**

### **Figure 1**

Infarct sizes (expressed as % of the area at risk) in different groups of anesthetized rabbits submitted to 30 min of coronary artery occlusion and 3 h of reperfusion. In the different groups, rabbits were either subjected to a normothermic protocol (Control group) or to extracorporeal blood cooling to 35°C or 32°C from the onset (0') or from the 10<sup>th</sup> or the 20<sup>th</sup> minute of ischemia (0', 10' and 20' ischemia). Open circles represent the individual infarct size of each animal and closed circles represent the mean±SEM of each group.

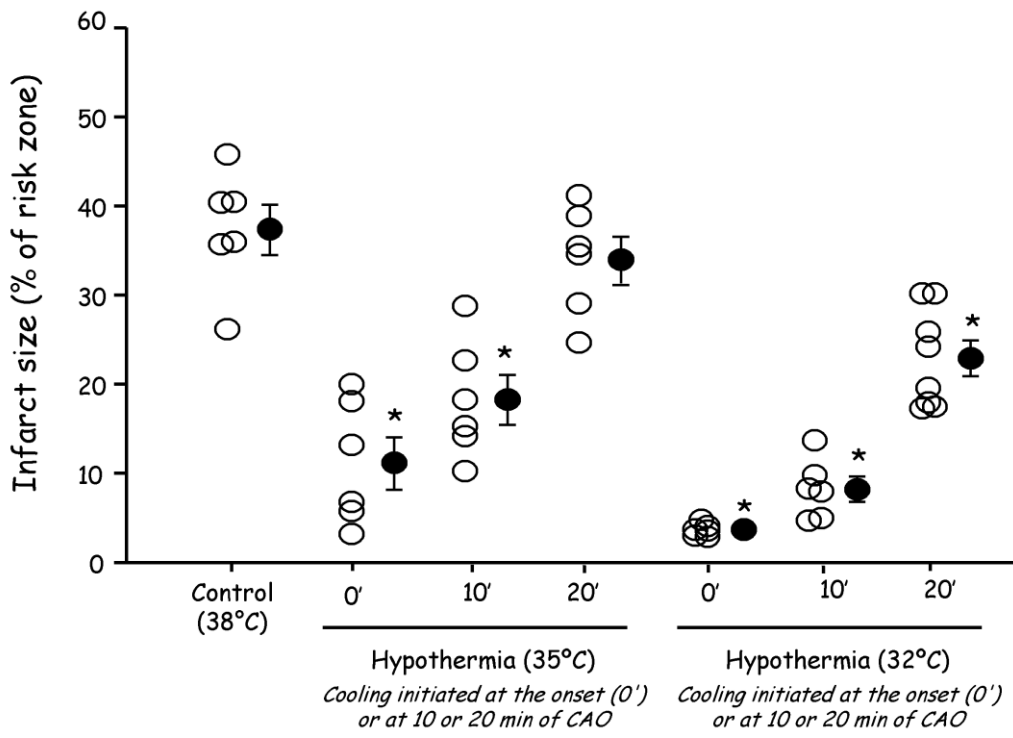
*Data adapted from Miki et al. <sup>12</sup> \*  $p < 0.05$  vs Control.*

### **Figure 2**

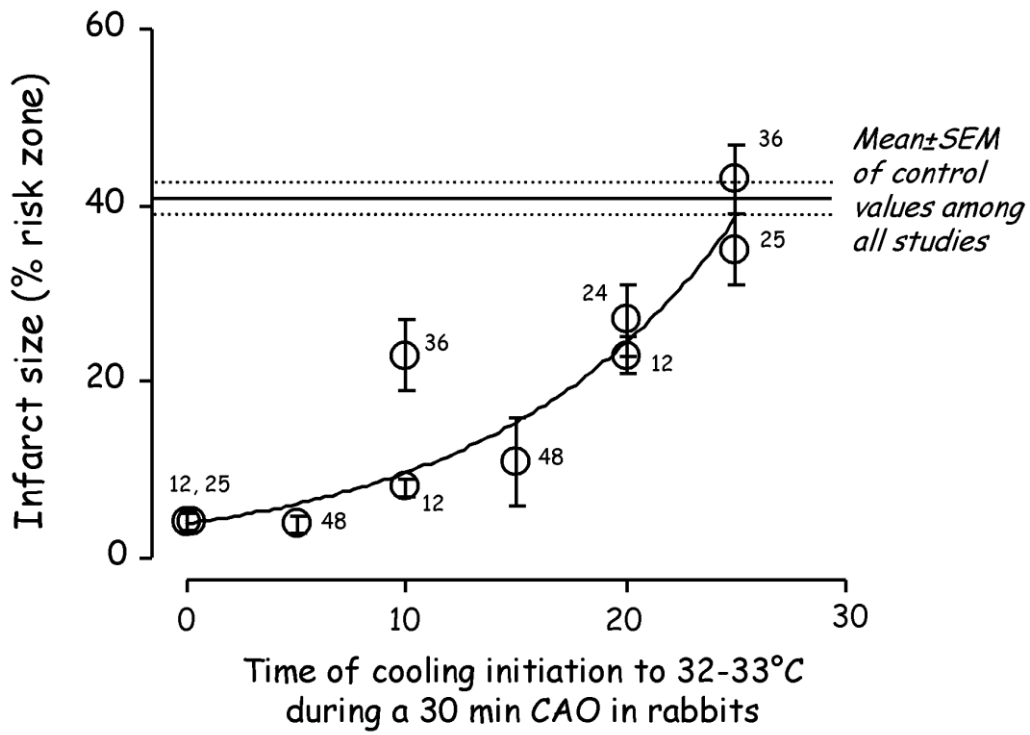
Infarct sizes (expressed as % of the area at risk) obtained from several studies in anesthetized rabbits subjected to 30 min of coronary artery occlusion and cooled to 32°C starting at different times after initiation of ischemia. Closed circles represent the mean±SEM of the cooled groups for each study.

*Numbers next to the data points are reference citations.*

**Figure 1**



**Figure 2**



**Table 1:** Summary of *in vivo* experimental studies investigating the infarct-limiting effect of myocardial cooling during ischemia, i.e., with cooling initiated before ischemia or at least 5 min before the end of coronary artery occlusion

| Species | Ref. | Cooling procedure                              | Duration of CAO (min) / CAR (h) | Target heart temperature | Time of cooling*                | IS with Cooling vs Control groups (% decrease) |
|---------|------|--|---------------------------------|--------------------------|---------------------------------|--|
| Rabbit  | 36   | Topical epicardial cooling                     | 30 min / 3 h                    | ~33°C                    | 10 min CAO → 15 min CAR         | 23±4 vs 44±4 (-48%)                            |
| Rabbit  | 22   | Topical epicardial cooling                     | 120 min / 3 h                   | ~30°C                    | 30 min CAO → 15 min CAR         | 59±3 vs 72±3 (-18%)                            |
| Rabbit  | 49   | Closed pericardioperfusion circuit             | 30 min / 3 h                    | ~34°C                    | -30 → 25 min CAO                | 18±3 vs 35±6 (-49%)                            |
| Rabbit  | 24   | Topical epicardial cooling                     | 30 min / 3 h                    | ~32°C                    | 20 min CAO → 120 min CAR        | 27±4 vs 51±5 (-47%)                            |
| Rabbit  | 12   | Blood cooling through heat exchanger           | 30 min / 3 h                    | ~35°C                    | 0 → 30 min CAO                  | 11±3 vs 37±3 (-70%)                            |
|         |      |  |                                 |                          | 10 → 30 min CAO                 | 18±3 vs 37±3 (-51%)                            |
|         |      |  |                                 |                          | 20 → 30 min CAO                 | 34±2 vs 37±3 (NS)                              |
|         |      |  |                                 |                          | 0 → 30 min CAO                  | 4±1 vs 37±3 (-89%)                             |
|         |      |  |                                 |                          | 10 → 30 min CAO                 | 8±1 vs 37±3 (-78%)                             |
| Rabbit  | 25   | Total liquid ventilation                       | 30 min / 3 h                    | ~32°C                    | 0 → 30 min CAO                  | 4±1 vs 38±1 (-89%)                             |
|         |      |  |                                 |                          | 5 → 30 min CAO                  | 4±1 vs 39±2% (-90%)                            |
| Rabbit  | 48   | Total liquid ventilation                       | 30 min / 72 h                   | ~32°C                    | 15 → 30 min CAO                 | 11±5 vs 39±2% (-72%)                           |
| Rabbit  | 37   | Surface cooling (water blankets)               | 30 min / 3 h                    | ~37.0°C                  | Before CAO → 180 min CAR        | 30±5 vs 59±1% (-48%)                           |
|         |      |  |                                 |                          | 0 min CAO → 180 min CAR         | 33±5 vs 59±1% (-43%)                           |
|         |      |  |                                 |                          | 15 min CAO → 180 min CAR        | 42±1 vs 59±1% (-28%)                           |
| Rabbit  | 52   | Topical epicardial cooling                     | 30 min / 3 h                    | ~35°C                    | -20 min before CAO → 15 min CAR | 16±3 vs 46±4 (-65%)                            |
| Rabbit  | 44   | Surface cooling                                | 60 min / 4 h                    | ~32°C                    | 5 → 30 min CAO                  | 78±10 vs 82±7 (NS)                             |
|         |      | Total liquid ventilation                       |                                 |                          | 5 → 30 min CAO                  | 45±18 vs 82±7 (-45%)                           |
|         |      |  |                                 |                          | 20 → 30 min CAO                 | 58±5 vs 82±7 (-29%)                            |
| Pig     | 26   | Endovascular cooling                           | 60 min / 3 h                    | ~34°C                    | 20 min CAO → 15 min CAR         | 9±6 vs 45±8 (-80%)                             |
| Pig     | 27   | Topical epicardial cooling                     | 40 min / 3 h                    | ~29°C                    | 0 → 40 min CAO                  | 25±2 vs 62±5 (-60%)                            |
| Pig     | 28   | Intracoronary cold saline infusion             | 60 min / 3 h                    | ~33°C                    | 15 min CAO → 15 min CAR         | 9±2 vs 36±4 (-75%)                             |
| Pig     | 35   | I.V. cold saline + endovascular cooling        | 40 min / ~4.3h                  | ~33°C                    | 25 min CAO → 25 min CAR         | 46±8 vs 75±5 (-39%)                            |
| Sheep   | 30   | Surface cooling (ice bags)                     | 60 min / 3 h                    | ~38.5°C                  | 0 min CAO                       | 63±2 vs 72±3 (-12%)                            |
|         |      |  |                                 | ~37.5°C                  | → 180 min CAR                   | 49±1 vs 72±3 (-31%)                            |
|         |      |  |                                 | ~36.5°C                  | Control Group at 39.5°C         | 39±1 vs 72±3 (-46%)                            |
|         |      |  |                                 | ~35.5°C                  |                                 | 22±2 vs 72±3 (-70%)                            |
| Dog     | 32   | Hypothermic retroperfusion of autologous blood | 210 min / 3 h                   | ~28-30°C                 | 30 → 210 min CAO                | 6±3 vs 24±7† (-75%)                            |

CAO, coronary artery occlusion; CAR, coronary artery reperfusion; IS, infarct size (expressed as % of area at risk); ref, reference number



*\*represents the time of the application of the cooling strategy and not the actual time at which the target temperature was reached. In several studies a delay was inevitable between the onset of the cooling protocol and the time of achievement of the target temperature (e.g., with low rate cooling strategy such as surface cooling).*

*† the Control value corresponds to infarct sizes observed with normothermic retroperfusion.*

**Table II:** Summary of *in vivo* experimental studies investigating the infarct-limiting effect of myocardial cooling during the reperfusion phase, i.e., with cooling started only 5 min before reperfusion or later yielding normothermic ischemia and hypothermic reperfusion

| Species | Ref. | Cooling procedure  | Duration of CAO (min) / CAR (h) | Target heart temperature | Time of cooling*         | IS with Cooling vs Control groups (% decrease) |
|---------|------|--|---------------------------------|--------------------------|--------------------------|--|
| Rabbit  | 36   | Topical epicardial cooling                                 | 30 min / 3 h                    | ~33°C                    | 25 min CAO → 15 min CAR  | 43±4 vs 44±4 (NS)                              |
| Rabbit  | 25   | Total liquid ventilation                                   | 30 min / 3 h                    | ~32°C                    | 25 min CAO → 30 min CAR  | 35±4 vs 38±1 (NS)                              |
| Rabbit  | 37   | Surface cooling (water blankets)                           | 30 min / 3 h                    | ~37.0°C                  | 25 min CAO → 180 min CAR | 44±2 vs 59±1% (-25%)                           |
|         |      |  |                                 |                          | 30 min CAO → 180 min CAR | 51±2 vs 59±1% (NS)                             |
| Pig     | 38   | Regional blood cooling through heat exchanger              | 45 min / 3 h                    | ~33°C                    | 43 min CAO → 120 min CAR | 71±8 vs 68±1 (NS)                              |
| Pig     | 28   | Intracoronary cold saline infusion                         | 60 min / 3 h                    | ~33°C                    | 0 min CAR → 30 min CAR   | 33±2 vs 45±5 (NS)                              |
| Pig     | 35   | Intravenous infusion of cold saline + endovascular cooling | 40 min / ~4.3h                  | ~33°C                    | 0 min CAR → 30 min CAR   | 80±6 vs 75±5 (NS)                              |

See Table 1 for legend.

**Table III:** Infarct size assessed by single photon emission computed tomography in patients presenting with ST-segment elevation myocardial infarction and treated by revascularization alone or in conjunction with endovascular cooling in the COOL-MI <sup>8</sup> and the ICE-IT <sup>10</sup> studies. Data were obtained from ref <sup>83</sup>, <sup>41</sup> and <sup>84</sup>.

|  | <b>Infarct size (% left ventricle)</b> |                    | <b><u>P</u><br/>value</b> |
|--|--|--------------------|---------------------------|
|  | Cooled group                           | Normothermic group |                           |
| <b><u>COOL-MI (n=392 patients)</u></b>                                       |  |                    |                           |
| Overall population   | 13.8%                                  | 14.1%              | 0.86                      |
| Subgroup with anterior STEMI cooled to <35° at the time of revascularization | 9.3%                                   | 18.2%              | 0.05                      |
| <b><u>ICE-IT (n=228 patients)</u></b>  |  |                    |                           |
| Overall population   | 10.2%                                  | 13.2%              | 0.14                      |
| Subgroup with anterior STEMI cooled to <35° at the time of revascularization | 12.9%                                  | 22.7%              | 0.09                      |

*STEMI, ST-segment elevation myocardial infarction*