

Hypothermia: a Therapeutic Option in Management of Stroke?

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Abstract

With an availability of a single medical therapy, ischaemic stroke continues to be one of the leading causes of mortality and morbidity in the World. Hence it is of paramount importance to find alternative therapies that can simultaneously affect various molecular mechanisms and demonstrate long term effects. Hypothermia has long been considered as a therapeutic option in preventing ischaemia-mediated brain damage and is already in place as an efficacious neuroprotective treatment regimen in neonatal hypoxic encephalopathy and in adults after cardiac arrest. However, before becoming an approved treatment for stroke, a comprehensive understanding of molecular mechanisms involved in its putative beneficial effects during or after ischaemic cerebral damage is needed. Furthermore, strong evidence on the optimal conditions in that hypothermic therapy may be administered and a thorough understanding of the associated potential complications are also required. This review paper initially discusses the optimal conditions in which hypothermic therapy may be safely applied before pointing out the systematic complications that may emerge from this treatment.

Keywords: Stroke; Ischaemia; Reperfusion; Therapeutic hypothermia; Rewarming; Shivering; Infection

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Introduction

Globally, stroke continues to be one of the leading causes of mortality and accounts for nearly 12% of total deaths every year [1-3]. It constitutes the leading cause of long term disability with up to 40% of stroke sufferers not recovering their independence and is one of the leading causes of disease burden when measured in disability adjusted life years [2,4]. Stroke also continues to be one of the most expensive conditions in that it costs National Health System £9 billion per annum in the UK alone [5].

There are two main types of stroke; ischaemic and haemorrhagic. The former constitutes about 85% of all strokes and can be further divided into two main subtypes; thrombotic and embolic strokes. Thrombotic strokes occur when a thrombus, formed in an atherosclerotic artery, occludes the blood flow to the distal part of this artery. This is usually preceded by a transient ischaemic attack or mini-stroke [6]. Embolic ischaemic strokes, on the other hand, usually arise when a blood clot breaks loose (embolus) and travels to a part of the cerebral vasculature that is too small to let it pass thereby significantly minimising or blocking the blood supply to the brain region supplied by this artery. In most cases the root-cause is cardioembolic [7].

Haemorrhagic strokes occur when cerebral blood vessels leading to or within the brain are ruptured or leak. Naturally, the damage from a haemorrhagic stroke are often more severe than ischaemic stroke. Moreover, haemorrhagic strokes have a higher risk of mortality than ischaemic strokes in the first 3 months. Similar to ischaemic strokes, there are two main types of haemorrhagic strokes which are determined by the location of the ruptured cerebral artery i.e. deep within the brain parenchyma (intracerebral haemorrhage) or on the surface of brain (subarachnoid haemorrhage) [8,9]. Since thrombolysis would increase the risk of further bleeding, it is contraindicated in the treatment of haemorrhagic stroke. Hence, the focus of medical treatment remains on gradual lowering of the blood pressure and intracranial pressure [9].

Current Treatment

To date, thrombolysis with recombinant tissue-plasminogen activator (r-tPA) remains as the only medical treatment option for ischaemic stroke. This thrombolytic agent converts the precursor

plasminogen into its active form, plasmin which degrades the fibrin that make up blood clots, leading to reperfusion of the ischaemic zone created by vascular occlusion [10-12]. However, r-tPA can only be given safely within the first 4.5 h of an ischaemic attack [12,13], hence many patients do not receive this treatment. Indeed, an audit by the Royal College of Physicians report that only 11.5% of all stroke patients between July 2014 and June 2015 were thrombolysed. The small time window for r-tPA administration, limited availability of stroke specialist centres due to resource constraints, shortage of specialised stroke physicians and the risk of haemorrhagic transformation may somewhat explain this [14,15]. Taken together, these findings indicate an obvious need for new therapies that may potentially mitigate the deleterious effects of debilitating condition [16,17].

Therapeutic Hypothermia

Due to its neuroprotective properties, hypothermia has long been considered as an important therapeutic strategy in the management of patients with hypoxic brain injury after cardiac arrest and in children with hypoxic-ischaemic encephalopathy [18]. Therapeutic hypothermia, defined as a core body temperature below 35°C, has later been shown to improve neurological integrity in cases of traumatic brain and spinal cord injuries [19]. Post-ischaemic suppression of a series of molecular and cellular mechanisms including oxidative stress, inflammation, neuroexcitotoxicity, blood-brain barrier disruption and apoptosis may account for the protective effects of therapeutic hypothermia in the central nervous system [20]. The specific mechanisms by which hypothermia may regulate these specific pathways have been the focus of a recent review paper [21]. However, the efficacy of hypothermia in the treatment of stroke has so far mainly been confined to various laboratory studies and pre-clinical trials. At present, there is not strong, clear evidence for the routine use of therapeutic hypothermia in clinical settings, indicating an urgent need for multi-centred, controlled, randomised clinical trials to test its safety, efficacy and feasibility in large number of stroke patients. These trials would undoubtedly be of great help in ascertaining a standard protocol for delivery of this therapy [22,23]. The evidence currently known about the optimal conditions will be outlined below.

Time of Onset

Currently there is not an optimum window of time in which therapeutic hypothermia is recommended. However, as the process of ischaemia and reperfusion occurs over a specific time course there must be a time-dependant window in which this treatment may be more effective. Although animal studies reveal the benefits of implementing hypothermic therapy at the onset of ischaemic injury, this may not be possible in clinical settings [24]. However, other animal studies have shown the therapeutic value of extending hypothermic treatment in cases where the cooling was started during the early phases of reperfusion [25,26]. A systematic review investigating the efficacy of therapeutic hypothermia in the treatment of animal models of ischaemic stroke has concluded that efficacy was highest when treatment was initiated "before or during the onset of ischaemia, in temporary rather than permanent ischaemia models".

However the paper has also reported that there is not a clear time dependency after the onset of ischaemia and even with a delay in the initiation of treatment (between 90-180 minutes), there was about 37% reduction in infarct size [27]. Taken together, these findings necessitate the establishment of a therapeutic time window and suggest that caution needs to be taken when interpreting the results as there is an inverse correlation between study quality and the impact of hypothermia.

Optimal Temperature

Temperatures below 31°C have shown the highest efficacy in reducing infarct size, implying that the lower the temperature, the better the neuroprotection and outcome. Even so, significant reductions in infarct size (30%) has also been obtained with mild hypothermia, 35°C [27]. Interestingly, animal studies focusing on the correlation between variations in temperature and infarct size reveal conflicting results e.g. while one study demonstrating a direct relationship between the level of decrease in brain temperature and the severity of subsequent ischaemic damage, another study showed better outcome with milder hypothermia. In the former study, 60% reduction in infarct size was observed in male spontaneously hypertensive rats subjected to transient middle cerebral artery occlusion (tMCAO) and had their brain temperature cooled to 34°C. Further reduction of temperature to 29°C in these animals led to eradication of all visible infarcts [28]. In the latter study, 3 hour exposure of rats to 32°C two hours after MCA occlusion produced a greater efficacy and tolerance than post-ischaemic cooling to 27°C [29]. In general, few studies support the application of temperatures below 32°C due to appearance of severe complications like arrhythmias, hypokalaemia and infections [16].

The current guidelines for a neuroprotective target temperature in patients with a return of circulation after cardiac arrest are 32-34°C, this may also be a feasible target in patients with ischaemic stroke [30]. Using tMCAO rat models a recent study has systematically compared the most effective target temperature for hypothermia in relation to both long and short term functional outcome where 6 different study groups were maintained at an integer between 37 and 32°C. The cooling occurred over 4 hours, 90 minutes after middle cerebral artery occlusion. After 24 hours functional outcome was highest in the 33 and 34°C group compared to other temperatures, with the greatest functional outcome in the 34°C group. This U-shaped curve was also apparent when looking at infarct size and oedema formation which was smallest in the 33-34°C groups, these effects persisted to the endpoint at 5 days. It was concluded from these results that the optimal depth of therapeutic hypothermia in tMCAO rats should be 34°C [31]. An effective range of 33-35°C, classed as mild to moderate hypothermia, is therefore recommended. Nevertheless, further studies investigating the optimal temperature depth after focal ischaemia are required [16,32].

Duration of Cooling

Similar to the time window for hypothermia, the optimal duration of therapeutic hypothermia is also unknown [16]. Intriguingly, van der Worp *et al* have reported an inverse

relationship between the duration of hypothermia and effect size in animal models. Considering that neurovascular damage after an acute ischaemic attack occurs over hours to days, it is assumed that a longer duration of hypothermia may lead to markedly improved outcomes [33,34]. In support of this hypothesis, some experiments have already demonstrated better outcomes with the longer duration of cooling. For instance, exposure of patients with traumatic brain injury to long-term mild hypothermia, i.e. 5 days, was more effective in improving outcome compared to short-term treatments with mild hypothermia (2 days). Taken together with the time course for pathological events after ischaemic stroke, these findings may strengthen the case for treatments with longer periods of hypothermia [35,36]. Furthermore, greater infarct size reduction and better outcomes were found in Sprague-Dawley rats subjected to tMCAO and treated with longer (21 hours) versus shorter (3 hours) periods of hypothermia [37]. In this regard, a separate study concludes that treatment of rodents with longer periods of hypothermia (>4 hours) during an ischaemic attack was more effective in neutralising hippocampal cellular damage than those with shorter periods of hypothermia i.e. 2 hours [38]. As mentioned above, the length of time prior to initiation of hypothermic treatment and the duration of cooling afterwards appear to be interdependent in that when there is a longer time to onset of treatment, a longer duration of cooling is required whilst with shorter delays in treatment, a smaller cooling duration appears to be adequate [16,24,39-41].

Clinical trials investigating the use of mild hypothermia for cardiac arrest indicate that the treatment is more efficacious when maintained for 12 to 24 hours, hence the National Institute for Health and Care Excellence (NICE) guidelines recommend 12-24 hours application of hypothermia [30,42,43]. This time frame may be a good indicator of a potential target in ischaemic stroke patients. Kallmunzer and Kollmar suggest the use of a surrogate parameter, like an MRI scan or measurement of a serum biomarker revealing the extent of neuronal damage, to determine the target temperature or duration of hypothermia [32].

Duration of Rewarming

The optimal rewarming rate for hypothermia in stroke management is yet to be established. A tMCAO animal study looking at rewarming rates in rats after treatments with hypothermia found that the animals had longer rewarming period (over 2 hours) had a smaller total infarct volume than those had faster rewarming, about 20 minutes. Even so, neurological outcome assessed using a Neuroscore system looking at forelimb posture, grasping reflex and spontaneous movements appeared to be similar in both the rapidly and slowly rewarmed animals after 5 days compared to normothermic animals [44,45]. These findings were contradictory to those of a clinical study investigating the rewarming rate after hypothermic cardiopulmonary bypass which reported a greater cognitive outcome at 6 weeks in patients subjected to a slower rate of rewarming, defined by <2°C difference between nasopharyngeal and cardiopulmonary bypass perfusate temperature, compared to controls who had 4-6°C difference between nasopharyngeal and cardiopulmonary bypass perfusate temperature [46]. The argument for a slower rewarming period is also supported

by studies looking into the importance of rewarming rate in hypothermic control of intracranial pressure or ICP [24,47,48]. In patients with massive hemispheric infarcts, the risk of a rebound rise in ICP was associated with a decreased rewarming duration (<16 hours). This rebound rise in ICP may be due to the hypermetabolic response causing a “rewarming shock” where there is sudden vasodilation to counter the insufficient cerebral blood flow mismatching with the increased metabolic demand during rewarming post hypothermia [49]. During rewarming, potassium levels should be monitored carefully as hyperkalaemia can occur due to the extracellular release of potassium that was sequestered intracellularly during hypothermia [50,51]. The rewarming process should be done with heated blankets and should not be faster than 0.5°C per hour to avoid temperature overshoot, with shivering and hypotension under control [52-54]. Current recommendations suggest a rewarming period of between 6-24 hours, with 12 hours being potentially optimal to avoid any further complications of prolonged rewarming [16,48].

Hypothermia Combined with Reperfusion Therapy

Previous studies exploring the effectiveness of therapeutic hypothermia in temporary versus permanent rodent models of human ischaemic stroke have collectively shown the inability of hypothermia to reduce infarct volume in permanent models of MCAO compared to respective control groups while being effective in tMCAO models and reducing infarct volume by about 48% [24,55,56]. Taken together these studies suggest that therapeutic hypothermia may augment the efficacy of r-tPA to recanalise the occluded arteries. However, the interaction between the effects of hypothermia and thrombolytic pharmacotherapy is complicated in that hypothermia has been shown to evoke coagulopathies through a variety of mechanisms including platelet dysfunction and is therefore implicated in haemorrhagic transformation [22,57-59]. Contrary to this, another study examining the thrombolytic activity of r-tPA in the presence of hyperthermia has shown lower thrombolytic activity at cooler temperatures [60]. It is also possible that recanalisation may negate the cooling efforts of hypothermia by increasing blood flow to the reperfused area and accelerating the increase in temperature [22,59,60].

Despite failure of an early animal study to reveal any additional benefit of combining hypothermia with r-tPA over treatment with hypothermia alone in embolic stroke models, a subsequent study investigating the effects of combinatory therapy using MRI outcomes reported a trend towards better survival in rats that had undergone hypothermia with early or late (1 and 3 hours after thromboembolic occlusion, respectively) thrombolysis compared to those that had thrombolysis alone. Cerebral perfusion as viewed by perfusion weighted imaging was also shown to improve in animals subjected to combinatory therapy, proving that therapeutic hypothermia did not interfere with the enzymatic activity of intravenous r-tPA (alteplase). Even so, no significant difference was found in overall results between groups treated with r-tPA alone or in combination with hypothermia [61,62]. Feasibility and safety of combining hypothermia and thrombolysis in the treatment of ischaemic stroke were

demonstrated in The Intravenous Thrombolysis Plus Hypothermia for Acute Treatment of Ischaemic Stroke (ICTuS-L) trial which looked at the safety of treatment with hypothermia (33°C degree achieved by endovascular cooling) and intravenous r-tPA within 6 hours of stroke symptom onset. Despite increased rates of pneumonia in the hypothermia versus normothermia group, this was neither associated with a poorer outcome nor an increased risk in bleeding [63]. As continuation of ICTuS-L study, the ICTuS 2/3 studies investigate whether the combination of thrombolysis and hypothermia is superior to thrombolysis alone for the treatment of acute ischemic stroke and whether there is an increased risk of pneumonia in combinatory treatment group [64]. In addition to this, EuroHYP-1, a European multicentre, randomised, phase 3 clinical trial aiming to reduce body temperature to 34-35°C within six-hours after symptom onset with rapid intravenous infusion of refrigerated normal saline or a surface cooling technique and maintained for 24 h is also currently underway to compare functional outcome in ischaemic stroke patients who undergo therapeutic hypothermia plus alteplase against patients who receive alteplase alone [65]. It is noteworthy that despite using different methods of cooling and target temperatures in both EuroHYP-1 and ICTuS trials, patients are also treated with pethidine and buspirone to prevent shivering and discomfort.

Combining therapeutic hypothermia with caffeine (caffeine plus ethanol) has also generated promising results *in vivo*. Indeed, an early study had shown that caffeine and 10% ethanol reduced the infarct volume in transient ischaemic rat models by about 83%. Delayed intravenous treatment of these animals with caffeine (up to 120 minutes) after ischaemic onset also resulted in significant reductions in infarct size compared to the controls [66]. A follow up study then investigated the effects of caffeine on infarct volume in the presence of hypothermia. For this, rodent MCAO models were exposed to 180 minutes of reversible ischaemia followed by 3 days of reperfusion. In conclusion, pairing caffeine with therapeutic hypothermia resulted in statistically significant decreases in infarct volume when compared to the control (treated with saline fluid) or either single treatment groups [67]. In this context, a pilot study, with 20 patients, was performed to test the safety and feasibility of combining caffeine and hypothermia with r-tPA. Of the 20 patients, 18 patients reached target temperature via endovascular or surface cooling methods and 3 died of causes unrelated to caffeine administration. Although this study has concluded that combining caffeine with hypothermia in acute ischaemic stroke patients received intravenous rt-PA is feasible, further prospective placebo-controlled randomised studies are required to explore safety and to test the efficacy of caffeine and/or hypothermia [68].

Systematic Effects and Complications

Several complications and systematic effects of therapeutic hypothermia such as rebound hyperkalaemia during rewarming, unwanted shivering and the risk of haemorrhage have already been mentioned so far. In addition to these, other systematic effects including an increased risk of infection, cardiovascular, haematological and metabolic problems also need to be considered while administering hypothermic treatment.

Shivering

Usually, shivering takes place when the core body temperature goes down to 30-35.5°C. Other than the discomfort it brings to patients, shivering can also negate the cooling effects of hypothermia by concurrently increasing vasocontractility and metabolic rate to increase heat production, implying a prerequisite for its clinical management during treatment with hypothermia [16,69]. Early detection remains a key factor in the prevention of shivering and ensuing irreversible physiological changes. To this end, the Bedside Shivering Assessment Scale has been developed which is a 4 point scale that rates shivering as absent, mild, moderate or severe and is assessed by inspection and palpation of different areas including the neck, thorax, arms and legs [70]. In clinical settings the intravenous opioid meperidine and orally administered buspirone are commonly used to suppress shivering without sedation or respiratory insufficiency. The combination of both drugs can successfully lower shivering threshold to 33.4°C [71]. In addition to this medical approach, skin surface rewarming may also be implemented to suppress shivering and vasoconstriction given that the cutaneous temperature constitutes around 20% of total thermoregulatory input in the body [72]. Meperidine and skin surface rewarming were found to act synergistically to reduce the shivering threshold to below 34°C with only mild sedation and no observed respiratory problems thereby indicating the efficacy of combining physical and pharmacological methods to control shivering and associated complications [73].

Cardiovascular and Haematological Effects

As core body temperature drops to 32°C, the heart rate also drops to around 40-45 beats per minute. This is a normal physiological response and does not require active intervention as ventricular filling time is longer, leading to a positive inotropic effect [17,74]. The risk of arrhythmia is not increased until core body temperature drops below 30°C. Indeed, no increases were reported in the frequency of arrhythmias at 33°C in comatose patients who had survived a cardiac arrest, supporting the notion that mild hypothermia would not adversely affect the risk of arrhythmias [69,75]. Research have shown that increases in mean arterial pressure during therapeutic hypothermia induce peripheral vasoconstriction which in turn increases venous return and triggers the release of atrial natriuretic peptide while suppressing the release of anti-diuretic hormone. Naturally, these may lead to "cold diuresis" associated with hypovolaemia [76].

In addition to haemodynamic effects, hypothermia may also modulate a series of haemostatic changes that may promote coagulopathies. At temperatures below 35°C there may be a mild reduction in platelet count whilst below 33°C the synthesis of clotting enzymes and plasminogen activators are affected [50]. This could potentially increase the risk of haemorrhagic transformation in patients with ischaemic stroke [77]. Despite this the risk of bleeding is not a reason to withhold treatment with very mild hypothermia, as this does not affect coagulation more than normothermia even in patients with a high bleeding risk [20]. However, as at present there is limited information

as to how hypothermia may affect recanalisation rates, more investigation is needed to look at the haemostatic changes that occur during and after hypothermia.

Infections

A series of distinct pathways including attenuation of leucocyte migration and phagocytosis as well as an increased release of cytokines from anti-inflammatory T cells may contribute to hypothermia-induced suppression of the immune system [50,78]. Although the dampening of the immune response may be protective in some ways, this can also be detrimental as it increases the body's susceptibility to infections such as pneumonia as evidenced by the ICTuS-L trial which showed a significant difference between the rates of pneumonia in the hypothermic versus normothermic group [63]. Contrary to this, a Cochrane review focusing on the effects of cooling therapy in ischaemic stroke has found no statistically significant difference in infection rates between the treatment and control groups [79]. Since sedatives, muscle relaxants and mechanical ventilation, administered as an adjunct alongside therapeutic hypothermia, could also increase the chance of infection [80-82], limiting the depth and duration of cooling may be one way of combatting the risks of infection due to lesser requirements for sedation and mechanical ventilation. However, this may also diminish the neuroprotective effects of therapeutic hypothermia. Hence, it is vital to find an optimal treatment regimen to establish balance between achieving neuroprotection and minimising the risk of infection. Another way to mitigate the risk of infection may involve the prophylactic use of antibiotics and early monitoring for potential signs of infection like fever or increased levels of C-reactive protein. Considering that, these may not be discernible in hypothermic patients, a daily blood screen cultures may be needed to check for bacteraemia. Bearing an increased risk of infection in mind, extra care should be taken at all times with catheter insertion and in preventing bedsores [20,83].

Electrolyte Disorders

Whilst therapeutic hypothermia is associated with a decreased urine output in some studies, many other studies reveal a radically increased diuresis due to decreased solute reabsorption in the ascending limb of the loop of Henle [84,85]. Whilst there is no significant change in serum Na^+ levels during hypothermia, the excretion of Mg^{2+} , K^+ and phosphate is markedly elevated to which intracellular sequestration of K^+ may also contribute. Due to the anticipated depletion of these electrolytes during hypothermic treatment, some patients may be given supplements to maintain them in the normal range. This is of particular importance given that low levels of Mg^{2+} can exacerbate brain injury while low levels of K^+ is implicated in cardiac arrhythmias. Indeed, polymorphic ventricular tachycardia has been closely associated with hypokalaemia in resuscitated cardiac arrest patients with ventricular fibrillation [17,86]. However, considering the risk of

rebound hyperkalaemia during rewarming care must be taken while administering K^+ supplementation [51].

Hyperglycaemia

Hypothermia reduces insulin secretion from the pancreas and as a consequence render patients hyperglycaemic [87,88]. As hyperglycaemia is associated with increased rates of infection, renal failure and neuropathy, its control by exogenous insulin may be necessary. Most hypothermic patients require higher than the physiological levels of insulin in order to maintain glucose levels within normal range due to the insulin resistance that may develop during the treatment [89]. Even so, no significant differences have been observed in plasma glucose levels between hypothermic and normothermic rodents subjected to permanent MCAO or in hypothermic versus normothermic patients with traumatic brain injury [26,51].

Other Effects

Hypothermia can impair bowel function by promoting the delay of ileal and gastric emptying. Hence, enteral support should be delayed until gut motility returns back to normal [52,69]. Furthermore, as hypothermia is a depressant on organ physiology and function in general whilst also suppressing the metabolism and excretion of multiple drugs, it makes logical sense that many pharmacological interventions will have their effects exaggerated during induced body cooling [85]. The impact of hypothermia on drug metabolism is variable and dependant on the route of elimination. In general the kinetics of most enzymatic pathways is slowed down during hypothermia. Consequently, Phase 1 metabolism, the initial biotransformation of a drug, usually by CYP450 enzymes, and Phase 2 metabolism, the conjugation of the drug by one or more molecular groups in order for it to be excreted, are the two most likely phases of drug metabolism and elimination to be affected [90]. Many CYP450 enzymes have been shown to have their activity inhibited during hypothermia, with processes such as active secretion or absorption showing marked reductions in activity [91]. As therapeutic hypothermia reduces clearance of many drugs and alters their potency, dose adjustment is recommended [17].

Conclusion

Therapeutic hypothermia alone or in combination with mechanical or pharmacological thrombectomy has long been regarded as a promising therapeutic option for ischaemic stroke. Although evidence concerning the time, duration and depth of the cooling periods as well as the rewarming rate continue to accumulate, further evidence is desperately required to devise a standard protocol that maintains the balance between neurovascular homeostasis and complications. Further pre-clinical and clinical trial studies are also needed to ascertain true value of this treatment in the settings of acute ischaemic stroke.

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