Dose-Response: An International Journal

Volume 9 | Issue 2 Article 5

6-2011

POSTCONDITIONING HORMESIS PUT IN PERSPECTIVE: AN OVERVIEW OF EXPERIMENTAL AND CLINICAL STUDIES

FAC Wiegant
Utrecht University, The Netherlands

HAB Prins
Utrecht University, The Netherlands

R Van Wijk
Utrecht University, The Netherlands

Dose-Response, 9:209–224, 2011
Formerly Nonlinearity in Biology, Toxicology, and Medicine

ISSN: 1559-3258
DOI: 10.2203/dose-response.10-004.Wiegant

International Dose-Response Society
www.Dose-Response.org

POSTCONDITIONING HORMESIS PUT IN PERSPECTIVE: AN OVERVIEW OF EXPERIMENTAL AND CLINICAL STUDIES

F.A.C. Wiegant □ University College, Utrecht University, The Netherlands.; Faculty of Science, Department of Biology, Utrecht University, The Netherlands

H.A.B. Prins □ University College, Utrecht University, The Netherlands

R. Van Wijk □ Faculty of Science, Department of Biology, Utrecht University, The Netherlands; International Institute of Biophysics, Neuss, Germany

□ A beneficial effect of applying mild stress to cells or organisms, that were initially exposed to a high dose of stress, has been referred to as 'postconditioning hormesis'. The initial high dose of stress activates intrinsic self-recovery mechanisms. Modulation of these endogenous adaptation strategies by administration of a subsequent low dose of stress can confer effects that are beneficial to the biological system. Owing to its potentially therapeutic applications, postconditioning hormesis is subject to research in various scientific disciplines. This paper presents an overview of the dynamics of postconditioning hormesis and illustrates this phenomenon with a number of examples in experimental and clinical research.

Keywords: hormesis, postconditioning, postexposure, adaptive response, preconditioning.

INTRODUCTION

Adaptation of cells and organisms (in response) to particular types of environmental stress is a widely studied phenomenon. Different types of stress include, for example, allergens, pathogens, ischemia, exercise, heavy metals or psychologically induced stress. Remarkably, instead of causing a continuously linear increase or decrease in response with dose, most of these stresses seem to act in accordance with a non-linear dose-response relationship known as the biphasic response. It is characterized by a J-shaped or an inverted U-shaped dose response curve representing low dose stimulation and high dose inhibition and is indicated with the term hormesis (Calabrese and Baldwin 2002a; Calabrese *et al.* 2007).

The phenomenon of hormesis has frequently been reported using a different nomenclature: for example, dose-response relationships are explained according to the Yerkes-Dodson Law in experimental psychology; the term subsidy-gradient is used in the area of ecological analysis; functional antagonism is employed nearly exclusively in enzymology; the

Address correspondence to Dr. F.A.C. Wiegant, Faculty of Science, Department of Biology, Cellular Dynamics, Utrecht University, P.O.Box 80056, 3508 TB, Utrecht, The Netherlands; Phone: -31-30-2533972; Fax: -31-30-2513655; E-mail: f.a.c.wiegant@uu.nl

U-shaped response, albeit applied in a wider range of disciplines, is commonly used in epidemiology; adaptive response is a term classically employed in molecular biology and physiology; biphasic response is frequently used in pharmacology; hormesis, finally, is the term that is most established in the area of toxicology where it was first mentioned (Calabrese 2008a). What these phenomena have in common is that their response relates biphasically to particular doses of stress. Such dose-response relationships are qualitatively independent of the stress-inducing agent, the endpoint measured and the system under investigation (Agutter 2008). Since no detailed explanatory mechanism for this evolutionary conserved phenomenon currently exists, unification of the hormesis-nomenclature is thwarted. Owing to Calabrese and others the term hormesis has gained wider general acceptance and, importantly, has increasingly come into use in the different scientific disciplines mentioned above (Calabrese and Baldwin 1998, 2002b, 2003; Calabrese *et al.* 2007). With numerous publications in various scientific journals, Calabrese has significantly contributed to the general acceptance of this evolutionarily conserved mechanism in the scientific world. Recently, researchers from various scientific fields have agreed on the universality of the phenomena they are studying (i.e., adaptive response, resilience, preconditioning stimulation) and, as proposed by Calabrese and colleagues, have agreed to use the term hormesis (Calabrese *et al.* 2007).

"Evolutionarily conserved" refers to the fact that throughout evolution, a specific phenomenon and its underlying mechanism has generally enhanced survival of the host and has therefore been preserved. "Adaptive" relates to intrinsic self-recovery mechanisms up-regulated in response to exposure to stress and possibly over-activation of these processes as a means of coping with the unpredictability of exposure to subsequent (other) stresses. The biphasic dose-response pattern is now generally thought to reflect the activation of the adaptive capacity of biological systems in response to stress (Agutter 2008; Mattson 2008).

Within the framework of hormesis, different approaches are used to elicit an adaptive response. Commonly, a stimulating low dose of stress is administered which initiates compensatory biological processes. These mechanisms that have been conserved through evolution confer a protective effect against exposure to a subsequent severe stress. This phenomenon is known as *preconditioning* hormesis. Less conventional is the administration of a low dose of stress to enhance repair and recovery processes *after* exposure to a more severe stress (Calabrese *et al.* 2007; Calabrese 2008b). The latter phenomenon is termed *postconditioning* hormesis. 'Post' in this case refers to treatment with a low dose of stimulating stress *after* an initial treatment with a large dose of stress. 'Hormesis' is the operational term used, and 'conditioning' pertains to the conditioning element of hormesis (Calabrese *et al.* 2007). The term

postconditioning hormesis may be preceded by the type of inducing agent. For example, *ischemic* postconditioning hormesis which involves low doses of hypoxic stress following a myocardial infarction, or *chemical* postconditioning hormesis which involves exposure to low levels of a chemical toxicant following a previous exposure to a severe stress condition such as a heat shock or a large amount of toxic agent, etc. Depending on whether the low-dose stress administered during postexposure is of the same type of stress or is different from the initial high-dose stress, postconditioning can be classified as *homologous* (i.e., the subsequent stress is the same as the initial stress) or *heterologous* (i.e., the subsequent stress is not the same as the initial stress).

Postconditioning, as part of the phenomenon of hormesis, is the main focus of this review paper. The phenomenon of postconditioning hormesis is not yet widely accepted. In this respect, Agutter (2008) recently questioned the generality of this phenomenon: whether it was indeed observed in numerous different biological systems or whether its occurrence is more restricted. Moreover, it was questioned whether this phenomenon has been observed at the cellular level or only in the context of multicellular organisms such as humans.

In this overview, examples in various scientific fields will be described where it has been recognized that, in addition to the protective effect of mild stress against damage from a subsequent more severe stress (i.e., preconditioning hormesis), application of a mild stress following a more severe stress condition can also confer the beneficial effects of postconditioning hormesis (Jonas and Ives 2008; Ovelgönne *et al.* 1995; Van Wijk *et al.* 1994a; Wiegant *et al.* 1997; Zhao 2007). In first instance, an example is described of a molecular biology research program in which the beneficial effect of mild doses of stress (including toxic compounds) has been observed when applied to stressed cells within the post-conditioning framework (eg Van Wijk *et al.* 1994a; Wiegant *et al.* 1997, 1998, 1999). In addition, a possible explanatory model for postconditioning hormesis will be discussed. Finally, some examples of clinical studies are described in which application of mild stress following more severe stress or application of mild stress to specific pathologic conditions appear to result in promising beneficial effects.

POSTCONDITIONING HORMESIS; A MOLECULAR APPROACH

In their research program on the possible beneficial effect of postconditioning using mild stress conditions, Van Wijk and Wiegant (2006, 2010) utilized 'in vitro' cultured Reuber H35 rat hepatoma cells that were pre-treated with an initial large dose of arsenite, cadmium or heat shock stress, immediately followed by incubation with lower doses of the initial stressor. Their program was focused in particular on the pro-

teotoxic response of cells and the regulation of stress protein synthesis. Hightower (1991) was the first to coin the term 'proteotoxicity' to indicate the detrimental action of damaged and denatured proteins that had lost their normal shape and function. The detrimental action of these denatured proteins in cells and organism has increasingly been recognized as an important focus of interest in biomedical disciplines (Gregersen *et al.* 2006; Morimoto 2008). A variety of adverse conditions, such as: an enhanced temperature (heat shock), free radical stress, radiation and heavy metals, can induce damage to cellular proteins. In response to proteotoxicity, cells react with an upregulation of heat shock proteins (hsps). These proteins function as molecular chaperones that facilitate the folding of newly synthesized proteins and repair structurally damaged proteins. In this way, the hsps are involved in repair, recovery and defence of protein structure and function. There are different families of these chaperone proteins (known as hsp28, hsp60, hsp70, hsp90 and hsp100) (Ellis 2007). The presence of these stress proteins lead to an increase in cell survival capacity under threatening conditions. It has since been demonstrated that chaperones possess many active functions: they repair structural damages by forcefully disentangle aggregated proteins, unfold and refold them into 're-educated and born again' functional proteins (Csermely and Vigh 2007; Ellis 2007). It is of interest that different stress conditions are able to induce characteristic differences in the type and in the quantity of the various stress proteins (Wiegant *et al.* 1994; Ryan and Hightower 1996). The question on which the research program focused was whether a postconditioning effect can be demonstrated at the molecular level of stress protein synthesis and on changes in survival capacity. Both homologous and heterologous postconditioning effects were evaluated.

Homologous postconditioning hormesis

In first instance, post-exposure studies were carried out using the homologous strategy. Cell cultures that were first exposed to a harmful (high dose) stress condition were subsequently incubated with lower doses of the initial stressor. When cells were damaged with chemical compounds such as arsenite (100 or 300 μM) or cadmium (10 or 30 μM) and subsequently exposed to low dose conditions of arsenite (1-10μM) or cadmium (0.1-1.0 μM) respectively, both hsp synthesis as well as survival capacity were enhanced due to post-conditioning (Ovelgönne *et al.* 1995; Wiegant *et al.* 1997). A further increase in hsp synthesis is interpreted as a beneficial phenomenon, since more chaperones will be available to assist in post-stress recovery (Van Wijk and Wiegant 2006; Wiegant and Van Wijk 2010).

However, a relation was observed between the severity of the pre-exposure- and the post-exposure condition. The more severe the initial

stress condition, the smaller the concentration required for stimulating hsp induction and survival capacity. When the initial stress condition has been severe, small doses used in postconditioning can unexpectedly merge into the harmful range. In this respect, it is of importance to note that without high dose pretreatment, the low dose stress condition used in post-exposure studies did not induce a detectable synthesis of stress proteins (hsps) nor were they able to affect survival capacity.

To study the effect of homologous heat postconditioning, cells were first exposed to a heat shock at 42°C or 43.5°C and subsequently incubated at mild 'fever-like' temperatures. An enhanced survival capacity and an enhanced synthesis of stress proteins were observed when the initial heat shock was followed by a post-exposure to a lower hyperthermic (fever-like) temperature (Schamhart *et al.* 1992; Van Wijk *et al.* 1994a). Similar observations were reported by Delpino *et al.* (1992). Also in these heat postconditioning studies the effect of low dose post-exposure appeared to be related to the severity of the pre-exposure temperature. Whereas the fever-like temperature enhanced survival capacity and hsp synthesis when applied 'postconditionally' following 42°C, the same fever-like temperature depressed survival capacity and inhibited hsp synthesis when applied following exposure of cells to 43.5°C (Van Wijk *et al.* 1994a).

These observations suggest a window of limited breadth for low doses of stressors in which beneficial effects may occur upon postconditioning.

Heterologous postconditioning hormesis

An intriguing question relates to the specificity of the stimulation of survival capacity and of hsp synthesis by postconditioning. In other words: does an initial damaging condition only sensitizes a cell population to stimulation by small doses of the same stress condition, or are cells also stimulated by small doses of heterologous stressors? In this respect, it is of interest that hormetic conditioning has been discussed with reference to the homeopathic similia principle (Bellavite *et al.* 2006a, 2006b; Van Wijk *et al.* 1994b; Van Wijk and Wiegant 2010). Samuel Hahnemann (1755-1843), a German physician and chemist who founded the therapeutic doctrine of homeopathy did so on the basis of the similia principle. According to this therapeutic principle, the same substance that causes the disease can be used in low doses to treat the disease (Bellavite *et al.* 2006b). In first instance, Hahnemann used dilutions of compounds, whereas at a later stage, a specific preparatory process was introduced in which 'potencies' are prepared using successive steps of dilution and succussion. These potencies may be diluted to such an extent that no molecule of the original solution is present in the actual remedy. These ultramolecular potencies cause controversy since an explanation of their sug-

gested effect is still lacking. In the here described research program, no ultramolecular but normal dilutions were used. According to the similia principle, there exists a similitude between the symptoms induced by the medicine and the symptoms of natural disease (Bellavite *et al.* 2006b). From this point of view, to cure the disease, compounds or conditions associated with the occurrence of particular symptoms can be used as stimulators of intrinsic self-recovery which then facilitates the diseased biological system to return to homeostasis (Van Wijk and Wiegant 2006; Wiegant and Van Wijk 2010). At the cellular level, the pattern of induced stress proteins (both heat shock proteins [hsps] and glucose-regulated proteins [grps]) is stressor-specific and was considered in the research program of Van Wijk and Wiegant as cellular 'symptoms' and hence as the sole indication to direct research as to the choice of the low dose agent to analyze its ability to exert beneficial effects in the recovery process. In this way, the specificity of the similia principle was studied (Wiegant *et al.* 1996, 1998, 1999). Following an initial heat shock, post-conditioning included low doses of arsenite, several heavy metal ions (cadmium, mercury, lead and copper), two different oxidative stress conditions (menadion and diethyldithiocarbamate) and a mild (homologous) hyperthermic temperature. Different stressors were shown to induce stressor-characteristic patterns of hsps (Wiegant *et al.*1994). Due to a selective induction of specific stress proteins qualitative as well as quantitative differences in hsp induction were observed. An example of a qualitative difference was that lead (Pb) induces the glucose regulated proteins (grp74 and grp94), which are not induced by other heavy metals, heat stress or oxidative stress and only slightly by arsenite. Another example of a qualitative difference is that cadmium and ddtc do not induce hsp60 whereas heat shock does. An example of a quantitative difference is that a specific hsp (such as hsp28) is induced to a different degree by various stress conditions. The degree of similarity between the pattern of hsps induced by the pre-exposure condition and those of the low dose postconditioning treatment was established in first instance (Wiegant *et al.* 1999). Then, in a set of experiments, cell cultures that were pre-exposed to a heat shock were subsequently post-exposed to different low dose stress conditions.

It is of interest that postexposure of heat-shocked cells to different chemical stress conditions demonstrated a differential stimulation of both tolerance development and of heat shock protein synthesis (Wiegant *et al.* 1999). The degree of stimulation appeared to depend on the similarity of both the molecular and cellular effects of the stress conditions used as initial disturbance and the postexposure treatment (Wiegant *et al.* 1998, 1999). A more detailed elaboration of this research program with respect to the evaluation of the similia principle at the cellular level, has recently been described by Wiegant and Van Wijk (2010).

Hahnemann's *similia similibus curentur* ('let like be cured by like') was met with much criticism and ridicule when it was introduced in the early 19th century. Up until today, neither the similia principle nor homeopathy as a medical system have become incorporated in current Western mainstream medicine. The comparative research on the relation between postconditioning hormesis and the homeopathic similia principle seems to offer an interesting challenge for both basic science and homeopathy in the years to come.

Although postconditioning, as a part of hormesis, reflects the similia principle, this is not necessarily so for the entire field of hormesis. Hormesis may also include preconditioning hormesis wherein administration of mild stress to a healthy system stimulates various adaptive mechanisms resulting in a protective effect against exposure to a subsequent, larger stress. Therefore, the preconditioning part of hormesis might primarily pertain to a medical prophylactic strategy, whereas postexposure conditioning would then relate primarily to a therapeutic strategy.

POSTCONDITIONING HORMESIS: SALUTOGENESIS AND CLINICAL UTILITY

Exactly which mechanisms underlie the phenomenon of postconditioning hormesis remains elusive. Yet the adaptive response mechanisms it elicits provide a functional link to homeostasis, which is a universal biological concept (Calabrese and Baldwin 2002a). Postexposure conditioning, specifically, involves initial exposure to a large amount of stress, followed by exposure to a much lower dose of stress. The initial severe stress activates intrinsic self-recovery mechanisms of the biological system which, subsequently, can be positively modulated by administration of a low dose of stress. Due to the severity of the initial stress, compensation that would otherwise return the system to the initial, balanced state (homeostasis) may not be sufficient. The extent to which the system has been brought out of balance determines the rates that control the maintenance of the state of the system.

Therefore, both homeostasis and homeorhesis should be considered in the control of states (Stebbing 2003). Together these principles allow a biological system, in response to administration of a large amount of stress, to adjust its rate and oscillate towards a state that is optimal under its current circumstances (i.e. not necessarily the previous homeostatic state). Such a response is in line with the Law of Initial Value (LIV), as proposed by Joseph Wilder (1895-1976) in 1935. According to this law, "the direction of response of a body function to any agent depends to a large degree on the initial level of that function" (Wilder 1962). In postconditioning, exposure to the first severe stress affects the initial level of the system. This initial level is now different from that of a healthy biological system; rather, it has become unbal-

anced (i.e., diseased). With a corresponding alteration in homeorhesis, exposure to a subsequent low dose of stress will elicit responses that differ from those elicited by the same amount of low-dose stress in a balanced (i.e. healthy) biological system.

The homeostatic and homeorhetic adaptations, set in motion by exposure and postexposure to severe and a low dose of stress, respectively, are the processes that provide a link between a state of health (salutogenesis or origin of health) and a state of disease (pathogenesis or origin of disease). Salutogenesis is a stress orientated concept which is focused on resources to maintain and improve a movement towards health (Antonovsky 1979; Charlton 1996; Lindström and Eriksson 2005). Whereas pathogenesis is primarily concerned with the cause of disease, salutogenesis pertains to the generation of a state of well-being (Davies 2007). The interplay and shifting of balances between homeostasis, salutogenesis and pathogenesis facilitates maintenance of an optimal state in the face of exposure to various amounts of stress. Health is dependent on the balance between salutogenesis and pathogenesis maintained by homeostasis (Davies 2007). This balance, in turn, is affected by hormetic conditioning, such as pre- and postconditioning hormesis.

In an experimental setting, accurate assessment of postconditioning hormesis involves 1). a sufficient amount of doses in the right quantities, 2). adequate dose spacing and 3). enough points in time for reliable evaluation (Calabrese 2008b). Often, assessment of postexposure conditioning within a clinical framework is even more laborious and difficult to realize. For example, an accurate indication of the current state of a particular biological system under study can be difficult to obtain. Application of postexposure conditioning within a clinical context is further complicated by the narrow window within which the inducing agent may initiate protective effects. Outside the therapeutic window, the postconditioning stress may become harmful. In addition, effectiveness of the agent used to induce stress can alternate depending on the biological system addressed and may differ, at the interindividual level, even within one type of biological system (Thayer *et al.* 2005). Moreover, low-dosage treatment with a particular harmful agent may profit one individual but may inversely affect the population at large or other biological systems in its direct environment. Conversely, considering that the dosage of stress is low, its effect is likely to be influenced by factors from the environment (Thayer *et al.* 2005). Despite these complexities, promising discoveries indicating the beneficial effects of exposing a biological system to a low adapting dose after a severe stress condition are emerging from different scientific disciplines. The next section provides a number of examples that illustrate the clinical potential of postexposure conditioning.

POSTCONDITIONING HORMESIS: EXAMPLES FROM EXPERIMENTAL AND CLINICAL RESEARCH

Postconditioning and Immunologic diseases

In immunology, postexposure conditioning has been related to the therapeutic effects of treatment with adjuvants, allergens and pathogens. This suggests a general principle. Particular types of adjuvants for example, can be used to purposely infect rats thereby triggering systemic inflammation. This stress can be greatly reduced by a subsequent injection of that same adjuvant in small doses. In this example of homologous postconditioning hormesis, the therapeutic effect was long lasting and antigen-specific because intraperitoneal aspecific inflammation, induced by another adjuvant, did not prevent the disease (Bellavite *et al.* 2006b).

Allergen-specific immunotherapy reflects the principle of (homologous) postexposure conditioning hormesis. This therapy is based on injecting patients suffering from allergies with increasing amounts of the offending allergen in an attempt to reduce their level of sensitivity to allergens (Bellavite *et al.* 2006b). For example, a case study on a patient whose milk sensitivity was repeatedly confirmed by dietary challenge, both blind and non-blind, reported sublingual milk therapy to be a successful treatment (i.e., homologous postconditioning hormesis) (Rapp 1978). Rapp demonstrated that if the patient ingested excessive amounts of milk and dairy products, the additional sublingual therapy relieved the symptoms of the patient. Another study on specific immunotherapy suggested clinical benefits for allergic asthma. The double-blind placebo controlled trial indicated that immunotherapy modified the natural progression of asthma by improving lung function (Wang 2007).

In patients with a chronic viral infection, therapeutic vaccination (homologous postconditioning hormesis) is aimed at modulating the host immune response to become more effective against the particular infection (i.e., reduce the level of susceptibility to the infection). Depending on the virus, chronic infection has, presumably, modulated the host immune response to become less effective against the invading virus thereby promoting viral persistence (Sällberg *et al.* 2007). Injection with additional material is thought to boost the immune response. Already in the mid-1900's, the bacillus Calmette-Guérin (BCG) vaccine was used to treat established infection with tuberculosis (Plotkin 1999). Other chronic viral infections that are considered possible targets for therapeutic vaccination are the hepatitis B virus (HBV), the hepatitis C virus (HCV) and human immunodeficiency virus (HIV-1) (Sällberg *et al.* 2007), which can be considered as a homologous postconditioning hormetic strategy.

In 1985, Karpas and colleagues described the principle of passive immunotherapy. HIV infected individuals received infusions of inacti-

vated plasma collected from symptomless HIV infected individuals (Karpas *et al.* 1985). Two studies, based on monthly infusions of this HIV-positive inactivated plasma demonstrated a positive clinical effect (Jackson *et al.* 1988; Karpas *et al.* 1988). Another study demonstrated the benefit of passive immunotherapy based on transfusion every 14 days during a period of one year. This treatment resulted in a delay of the appearance of the first AIDS-defining event (Vittecoq *et al.* 1995). Since the findings of Karpas and colleagues, both clinical observations and systematic trials related to homologous postconditioning hormesis have suggested a correlation between a.) the goal of the immunotherapy, that is, to decrease viral loads and increase CD4+ cell counts, and b.) the decreased morbidity and mortality resulting from HIV-1 infection (Connolly *et al.* 2008).

Postconditioning and Cardiovascular disease

Ischemic preconditioning and ischemic postconditioning demonstrate consistent and clear cardioprotective effects utilized in every experimental animal model (Liem *et al.* 2007). For example, the homologous postexposure conditioning effect of repetitive ischemia applied during early reperfusion in anesthetized open-chest dogs was cardioprotective by attenuating reperfusion injury (Zhao *et al.* 2003). Postconditioning could be an efficient cardioprotective intervention in patients with acute myocardial infarction submitted to primary percutaneous coronary interventions (Darling *et al.* 2007; Laskey 2005; Staat *et al.* 2005). It has been suggested that postconditioning protection for ischemia/reperfusion-induced hypercontracture and cell death might be due to prolongation of intracellular acidosis during initial reperfusion (Inserte *et al.* 2008). Other studies have demonstrated that postconditioning can protect the kidney from ischemia/reperfusion injury. Presumably, the mechanism that involves protection of the mitochondrial respiratory chain function is similar to the heart model (Serviddio *et al.* 2008). Other research has indicated that postconditioning also reduces ischemic damage in the brain. However, it remains to be elucidated whether or not this type of postconditioning provides long-term protection and also improves neurological function (Zhao 2007; 2009) or in myocardial infarction (Skyschally *et al.* 2009).

Physical exercise is viewed as a potential non-pharmacological antihypertensive treatment for diet-induced obesity hypertension (Pinheiro *et al.* 2007). Low intensity exercise training has been reported to attenuate the decrease in $\dot{V}O_{2max}$ during the lifespan of a rat (Tipton *et al.* 1983) and also decreases blood pressure in the majority of patients with hypertension (Hagberg *et al.* 2000). In this example of postconditioning hormesis, a temporary, moderate increase in blood pressure, typically associated with exercise, results in improvement in

hypertensive patients (i.e., an eventual decrease in blood pressure). Research in both obese and non-obese rats supports the impression that hypertension, subsequent cardiac hypertrophy, myocardial fibrosis and myocardial vascularization improved in response to exercise training (Pinheiro *et al.* 2007).

Postconditioning and Cancer

Although harmful in high doses, radiation in low doses may stimulate various immune functions that have the potential for clinical application (Calabrese and Baldwin 2002b). For example, total-body irradiation (TBI) in high doses is commonly used prior to bone marrow transplantation due to its immunosuppressive effects, yet in low doses TBI has been shown to suppress metastasis in tumor-bearing rats through various immune-stimulating mechanisms (Hashimoto *et al.* 1999). Other research supports the notion that low doses of TBI to mice may delay tumor growth under certain conditions (Ito *et al.* 2008). In humans, low dose radiation has been introduced as an effective therapy of non-Hodgkin's lymphoma (Girinsky *et al.* 2001; Kennerdell *et al.* 1999; Richaud *et al.* 1998). Richaud and colleagues demonstrated that most patients treated with low dose radiation showed an efficient response, particularly with follicular lymphoma. In these patients, neither acute nonlymphoblastic leukemia nor myelodysplasic syndrome was observed with a median follow-up of 88 months.

Postconditioning and Neurological and Psychological diseases

Postconditioning hormesis has been therapeutically linked to treatment of brain injury and psychological disorders. For example, glutamate is the primary toxin released during brain injury. Several studies have shown that treatment with low-dose glutamate can be used to reduce brain injury after trauma (Ives *et al.* 2001; Jonas *et al.* 1999, 2001; Marotta *et al.* 2002) thereby reflecting postconditioning hormesis.

In psychology, postconditioning hormesis is recognized in exposure therapy for a wide variety of phobias. Exposure therapy is an effective means of reducing negative affective symptoms associated with different phobias (Rothbaum and Schwartz 2002). In recent experiments, phobic subjects showed increased activity in the amygdala, a key brain structure implicated in fear sensation. Two weeks after exposure therapy, a significant reduction in hyperactivity at this location was recorded (Goossens *et al.* 2007). These findings suggest a neural basis for modulating emotional experience through systematic exposure to phobic stimuli (Hariri *et al.* 2000). A recent development in the treatment of phobias is virtual reality exposure therapy (VRET), a novel tool that has been shown to reduce anxiety and phobia symptoms (Parsons and Rizzo 2008).

Postconditioning and Intoxication

Application of postconditioning hormesis also appears in the field of toxicology. In an early multi-centred, double-blind study, administration of low-dose mustard gas and an extract of the plant toxin *Rhus toxicodendron* was suggested to modify local mustard-induced blistering and hasten the rate of healing (Paterson 1944). Recently, research suggests that exposure to sub-toxic, low-levels of toxic agents may stimulate a sufficient protective response for both prevention *and* treatment of the effects resulting from exposure to higher doses of toxin. This may provide an alternative protective approach from biological and chemical agents (Jonas and Ives 2008; Szeto *et al.* 2004).

DISCUSSION AND CONCLUSION

The generally accepted evolutionarily conserved phenomenon of hormesis involving both low-dose stimulation and high-dose inhibition has "evolved" over decades in many fields. Initially, hormesis had only been identified with pre-exposure conditioning. Post-exposure conditioning has only recently arrived into the field of hormesis and has therefore not yet reached a comparable level of acceptance in the scientific world. Postconditioning hormesis involves administration of a low dose of stress following exposure to a severe stress. Recently, the generality of this phenomenon has been questioned, including whether beneficial effects could be observed at the cellular level (Agutter 2008). This paper presents a number of reports from both "in vitro" cellular research and current clinical research illustrating the relevance and therapeutic potential of postconditioning hormesis.

In the current overview, examples of cellular, experimental and clinical research are described from a phenomenological point of view. In this overview, the observed phenomena are not explained in detailed molecular or mechanistic terms. Although a number of papers have been mentioned that speculate on the molecular basis of hormesis (Agutter 2008; Mattson 2008; Stebbing 2003), further studies are required to understand the underlying mechanism of both pre- and post-conditioning hormesis. In this respect, it should be noted that an improved mechanistic understanding may require a recategorization of these phenomena in the future.

With respect to the possible underlying mechanism, it is speculated that the initial severe stress affects the homeostatic (balanced) state of a biological system and activates intrinsic self-recovery mechanisms. Administration of a subsequent low dose stress may modulate or over-activate these recovery mechanisms attempting to cope with possible but unpredictable encounters with (other) future stresses. Activation of these adaptation strategies with a low dose stress can confer beneficial effects to

the biological system. Exactly which mechanisms underlie this phenomenon remains elusive. However, the initial exposure alone, and/or combined with post-exposure, activate homeostatic and homeorhetic adaptations which provide a link between a state of health (salutogenesis) and a state of disease (pathogenesis). The shifting between these states facilitates maintenance of optimal balance in the face of future exposure to stress. Another aspect that requires further attention is both the concentration as well as the nature in which the low dose stimulus should be applied. With respect to the concentration, it has been shown that the benefit of the low dose effect depends on the actual sensitivity of the biological system (Ovelgönne *et al.* 1995; Wiegant *et al.* 1999). In addition, the ultramolecular dilutions that were shown to support recovery (Paterson 1944; Ives *et al.* 2001; Jonas *et al.* 1999; 2001) is an aspect that requires an explanation with respect to the nature of the information transfer leading to a modulation of recovery mechanisms. Although the underlying mechanisms in which recovery mechanisms are activated by low doses remain obscure, promising results indicating the beneficial effects of post-exposure conditioning are emerging from different scientific disciplines. Further research in this field represents an interesting challenge for both fundamental and clinical measurements.

REFERENCES

Agutter PS. 2008. Elucidating the mechanism(s) of hormesis at the cellular level: the universal cell response. Am J Pharmacol Toxicol 3: 97-107.

Antonovsky A. 1979. Health, stress and coping. Jossey-Bass, San Francisco.

Bellavite P, Conforti A, Pontarollo F and Ortolani R. 2006a. Immunology and homeopathy. 2. Cells of the immune system and inflammation. eCAM 3: 13-24.

Bellavite P, Ortolani R and Conforti A. 2006b. Immunology and homeopathy. 3. Experimental studies on animal models. eCAM 3: 171-186.

Calabrese EJ and Baldwin LA. 1998. Hormesis as a biological hypothesis. Env Health Persp 106: 357-362.

Calabrese EJ and Baldwin LA. 2002a. Defining hormesis. Hum Exp Toxicol 21: 91-97.

Calabrese EJ and Baldwin LA. 2002b. Applications of hormesis in toxicology, risk assessment and chemotherapeutics. Trends Pharm Sci 23: 331-337.

Calabrese EJ and Baldwin LA. 2003. Hormesis: the dose-response revolution. Annu Rev Pharmacol Toxicol 43: 175-197.

Calabrese EJ, Bachmann KA, Bailer AJ, Bolger PM, Borak J, Cai L, Cedergreen N, Cherian MG, Chiueh CC, Clarkson TW, Cook RR, Diamond DM, Doolittle DJ, Dorato MA, Duke SO, Feinendegen L, Gardner DE, Hart RW, Hastings KL, Hayes AW, Hoffmann GR, Ives JA, Jaworowsk, Z, Johnson TE, Jonas WB, Kaminski NE, Keller JG, Klaunig JE, Knudsen TB, Kozumbo WJ, Lettieri T, Liu S-Z, Maisseu A, Maynard KI, Masoro EJ, McClellan RO, Mehendale HM, Mothersill C, Newlin DB, Nigg HN, Oehme FW, Phalen RF, Philbert MA, Rattan SIS, Riviere JE, Rodricks J, Sapolsky RM, Scott BR, Seymour C, Sinclair DA, Smith-Sonneborn J, Snow ET, Spear L, Stevenson DE, Thomas Y, Tubiana M, Williams GM and Mattson MP. 2007. Biological stress response terminology: Integrating the concepts of adaptive response and preconditioning stress within a hormetic dose–response framework. Toxicol Appl Pharmacol 222: 122–128.

Calabrese EJ. 2008a. Converging concepts: Adaptive response, preconditioning, and the Yerkes-Dodson law are manifestations of hormesis. Ageing Res Rev 7: 8-20.

Calabrese EJ. 2008b. Hormesis: Why is it important to toxicology and toxicologists. Env Toxicol Chem 27: 1451-1474.

Charlton BG. 1996. A new science of health: salutology and the evolutionary perspective. Q J Med 89: 233-236.

Connolly NC, Whiteside TL, Wilson S, Kondragunta V, Rinaldo CR and Riddler SA. 2008. Therapeutic immunization with human immunodeficiency virus type 1 (HIV-1) peptide-loaded dendritic cells is safe and induces immunogenicity in HIV-1-infected individuals. Clin Vaccine Immunol 15: 284-292.

Csermely P and Vigh L (eds). 2007. Molecular Aspects of the Stress Response: Chaperones, Membranes and Networks. Landes Bioscience and Springer Science+Business media, New York.

Darling CE, Solari PB, Smith CS, Furman MI and Przyklenk K. 2007. Postconditioning the human heart: Multiple balloon inflations during primary angioplasty may confer cardioprotection. Basic Res Cardiol 102: 274-278.

Davies PG. 2007. Between health and illness. Persp Biol Med 50: 444-452.

Delpino A, Gentile FP, Di Modugno F, Benassi M, Mileo AM and Mattei E. 1992. Thermo-sensitization, heat shock protein synthesis and development of thermotolerance in M-14 human tumor cells subjected to step-down heating. Radiat Environ Biophys 31: 323-332.

Ellis RJ. 2007. Protein misassembly: macromolecular crowding and molecular chaperones. In: Molecular Aspects of the Stress Response: Chaperones, Membranes and Networks. (P. Csermely and L. Vigh (eds.)). Landes Bioscience and Springer Science+Business Media. New York. pp1-13.

Girinsky T, Guillot-Vals D, Koscielny S, Cosset JM, Ganem G, Carde P, Monhonval M, Pereira R, Bosq J, Ribrag V, Vantelon JM, Munck JN. 2001. A high and sustained response rate in refractory or relapsing low-grade lymphoma masses after low-dose radiation: analysis of predictive parameters of response to treatment. Int J Radiat Oncol Biol Phys 51: 148-155.

Goossens L, Sunaert S, Peeters R, Griez EJL, Schruers KRJ. 2007. Amygdala hyperfunction in phobic fear normalizes after exposure. Biol Psych 62: 1119-1125.

Gregersen N, Bross P, Vang S and Christensen JH. 2006. Protein misfolding and human disease. Annu Rev Genomics Hum Genet 7: 103-124.

Hagberg JM, Park J-J and Brown MD. 2000. The role of exercise training in the treatment of hypertension: an update. Sports Med 30: 193-206.

Hariri AR, Bookheimer SY and Mazziotta JC. 2000. Modulating emotional responses: Effects of a neocortical network on the limbic system. Neuroreport 11: 43-48.

Hashimoto S, Shirato H, Hosokawa M, Nishioka T, Kuramitsu Y, Matushita K, Kobayashi M and Miyasaka K. 1999. The suppression of metastases and the change in host immune response after low-dose total-body irradiation in tumor-bearing rats. Rad Res 151: 717-724.

Hightower LE. 1991. Heat shock, stress proteins, chaperones, and proteotoxicity. Cell 66: 191-197.

Inserte J, Barba I, Hernando V, Abellán A, Ruiz-Meana M, Rodríguez-Sinovas A and Garcia-Dorado D. 2008. Effect of acidic reperfusion on prolongation of intracellular acidosis and myocardial salvage. Cardiovasc Res 77: 782-790.

Ito M, Shibamoto Y, Avakwa S, Tomita N, Sugie C and Ogino H. 2008. Effect of low-dose total-body irradiation on transplantability of tumor cells in syngeneic mice. J Rad Res 49: 197-201.

Ives J, Jonas W, Hartings J. *et al.* 2001. Neuroprotection in experimental rat brain injury with ultra high dilution glutamate but not potassium chloride. Soc Neurosci Abstr 27: 557.

Jackson GG, Perkins JT, Rubenis M, Paul DA, Knigge M, Despotes JC and Spencer P. 1988. Passive immunoneutralization of human immunodeficiency virus in patients with advanced AIDS. Lancet 2(8612): 647-652.

Jonas WB, Lin Y, Williams A, Tortella F and Tuma R. 1999. Treatment of experimental stroke with low-dose glutamate and homeopathic Arnica Montana. Perfusion 12, 452-462.

Jonas WB, Lin Y and Tortella F. 2001. Neuroprotection from glutamate toxicity with ultra-low dose glutamate. Neuro Reports 12: 335-339.

Jonas WB and Ives JA. 2008. Should we explore the clinical utility of hormesis? Hum Exp Toxicol 27: 123-127.

Karpas A, Gillson W, Bevan PC and Oates JK. 1985. Lytic infection by British AIDS virus and development of rapid cell test for antiviral antibodies. Lancet 2(8457): 695-697.

Karpas A, Hill F, Youle M, Cullen V, Gray J, Byron N, Hayhoe F, Tenant-Flowers M, Howard L, Gilgen D, Oates JK, Hawkins D and Gazzard B.1988. Effects of passive immunization in patients with the acquired immunodeficiency syndrome-related complex and acquired immunodeficiency syndrome. Proc Natl Acad Sci USA 85: 9234-9239.

Kennerdell JS, Flores NE and Hartsock RJ. 1999. Low-dose radiotherapy for lymphoid lesions of the orbit and ocular adnexa. Ophthal Plast Reconstr Surg 15: 129-133.

Laskey WK. 2005. Brief repetitive balloon occlusions enhance reperfusion during percutaneous coronary intervention for acute myocardial infarction: a pilot study. Catheter Cardiovasc Interv 65: 361-367.

Liem DA, Honda HM, Zhang J, Woo D and Ping P. 2007. Past and present course of cardioprotection against ischemia-reperfusion injury. J Appl Physiol 103: 2129-2136.

Lindström B and Eriksson M. 2005. Salutogenesis. J Epidemiol Comm Health 59: 440-442.

Marotta D, Marini A, Banaudha K, Maharaj SV and Jonas WB. 2002. Non-linear effects of cycloheximide in glutamate treated cultured rat cerebellar neurons. Neurotoxicol 23: 307-312.

Mattson MP. 2008. Hormesis and disease resistance: activation of cellular stress response pathways. Hum Exp Toxicol 27: 155-162.

Morimoto RI. 2008. Proteotoxic stress and inducible chaperone networks in neurodegenerative disease and aging. Genes Dev 22: 1427-1438.

Ovelgönne JH, Wiegant FAC, Souren JEM, Van Rijn J and Van Wijk R. 1995. Enhancement of the stress response by low concentrations of arsenite in arsenite-pretreated H35 hepatoma cells. Toxicol Appl Pharmacol 132: 146-155.

Parsons TD and Rizzo AA. 2008. Affective outcomes of virtual reality exposure therapy for anxiety and specific phobias: a meta-analysis. J Behav Ther Exp Psychiatry 39: 250-261.

Paterson J. 1944. Report on mustard gas experiments. J Am Homoeop Assoc 37: 47-50 / 37: 88-89.

Pinheiro AR, Cunha AR, Aguila MB and Mandarim-de-Lacerda CA. 2007. Beneficial effects of physical exercise on hypertension and cardiovascular adverse remodeling of diet-induced obese rats. Nutr Metab Cardiovasc Res 17: 365-375.

Plotkin SA. 1999. Vaccination against the major infectious diseases. C R Acad Sci III 322: 943-951.

Rapp DJ. 1978. Double-blind confirmation and treatment of milk sensitivity. Med J Aust 1: 571-572.

Richaud PM, Soubeyran P, Eghbali H, Chacon B, Marit G, Brouset A and Hoerni B. 1998. Place of low-dose total body irradiation in the treatment of localized follicular non-Hodgkin's lymphoma: results of a pilot study. Int J Radiat Oncol Biol Phys 40: 387-390.

Rothbaum BO and Schwartz AC. 2002. Exposure therapy for posttraumatic stress disorder. Am J Psychother 56: 59-75.

Ryan JA and Hightower LE. 1996. Stress proteins as molecular biomarkers for environmental toxicology. EXS. 77: 411-424.

Sällberg M, Weiland M and Frelin L. 2007. Therapeutic vaccines: challenges of chronic viral infections. Drug Disc Today: Therap Strateg 4: 253-266.

Schamhart DHJ, Zoutewelle G, Van Aken H and Van Wijk R. 1992. Effects on the expression of heat shock proteins by step-down heating and hypothermia in rat hepatoma cells with a different degree of heat sensitivity. Int J Hyperthermia 8: 701-716.

Serviddio G, Romano AD, Gesualdo L, Tamborra R, Di Palma AM, Rollo T, Altomare E and Vendemiale G. 2008. Postconditioning is an effective strategy to reduce renal ischemia/reperfusion injury. Nephrol Dial Transpl 23: 1504-1512.

Skyschally A, Van Caster P, Iliodromitis EK, Schulz R, Kremastinos DT and Heusch G. 2009. Ischemic postconditioning: experimental models and protocol algorithms. Basic Res Cardiol 104: 469-483.

Staat P, Rioufol G, Piot C, Cottin Y, Cung TT and L'Huillier I. *et al.* 2005. Postconditioning the human heart. Circulation 112: 2143-2148.

Stebbing ARD. 2003. A mechanism for hormesis – a problem in the wrong discipline. Crit Rev Toxicol 33: 463-467.

Szeto AL, Rollwagen F and Jonas WB. 2004. Rapid induction of protective tolerance to potential terrorist agents: a systematic review of low- and ultra-low dose research. Homeopathy 93: 173-178.

Thayer KA, Melnick R, Burns K, Davis D and Huff J. 2005. Fundamental flaws of hormesis for public health decisions. Env Health Persp 113: 1271-1275.

Tipton CM, Matthes RD, Marcus KD, Rowlett KA and Leininger JR. 1983. Influences of exercise intensity, age, and medication on resting systolic blood pressure of SHR populations. J Appl Physiol 55: 1305-1310.

Van Wijk R, Ovelgönne JH, de Koning E, Jaarsveld K, Van Rijn J, Wiegant FAC. 1994a. Mild step-down heating causes increased transcription levels of hsp68 and hsp84 mRNA and enhances thermotolerance development in Reuber H35 hepatoma cells. Int J Hyperthermia 10: 115-125.

Van Wijk R, Ooms H, Wiegant FAC, Souren JEM, Ovelgönne JH, Van Aken JM, and Bol AWJM. 1994b. A molecular basis for understanding the benefits from subharmful doses of toxicants. Environm Management and Health 5: 13-25.

Van Wijk R and Wiegant FAC. 2006. The Simila Principle. An Experimental Approach on the Cornerstone of Homeopathy. Karl und Veronica Carstens-Stiftung, Essen.

Van Wijk R and Wiegant FAC. 2010. Post-conditioning hormesis and the homepathic similia principle: molecular aspects. Human Exp Toxicol 29: 561-565.

Vittecoq D, Chevret S, Morand-Joubert L, Heshmati F, Audat F, Bary M, Dusautoir T, Bismuth A, Viard JP and Barré-Sinoussi F. 1995. Passive immunotherapy in AIDS: a double-blind randomized study based on transfusions of plasma rich in anti-human immunodeficiency virus 1 antibodies vs. transfusions of seronegative plasma. Proc Natl Acad Sci USA 92: 1195-1199.

Wang J. 2007. Allergy immunotherapy as an early intervention in patients with child-onset atopic asthma. Pediatrics 120: S148-S149.

Wiegant FAC, Souren JEM, Van Rijn J and Van Wijk R. 1994. Stressor-specific induction of heat shock proteins in rat hepatoma cells. Toxicology 94: 143-159.

Wiegant FAC, Spieker N, Van der Mast CA and Van Wijk R. 1996. Is heat shock protein re-induction during tolerance related to the stressor-specific induction of heat shock proteins? J Cell Physiol 169: 364-372.

Wiegant FAC, Van Rijn J and Van Wijk R. 1997. Enhancement of the stress response by minute amounts of cadmium in sensitized Reuber H35 hepatoma cells. Toxicology 116: 27-37.

Wiegant FAC, Spieker N and Van Wijk R. 1998. Stressor-specific enhancement of hsp induction by low doses of stressors in conditions of self- and cross-sensitization. Toxicology 127: 107-119.

Wiegant FAC, Souren JEM and Van Wijk R. 1999. Stimulation of survival capacity in heat-shocked cells by subsequent exposure to minute amounts of chemical stressors: Role of similarity in hsp-inducing effects. Hum Exp Toxicol 18: 460-470.

Wiegant FAC and Van Wijk R. 2010. The similia principle; results obtained in a cellular model system. Homeopathy 99: 3-14.

Wilder J. 1962. Basimetric approach (law of initial value) to biological rhythms. Ann NY Acad Sci 30: 1211-1220.

Zhao, ZQ, Corvera JS, Halkos ME, Kerendi F, Wang NP, Guyton RA and Vinten-Johansen J. 2003. Inhibition of myocardial injury by ischemic postconditioning during reperfusion: comparison with ischemic preconditioning. Am J Physiol Heart Circ Physiol 285: H579-H588.

Zhao H. 2007. The protective effect of ischemic postconditioning against ischemic injury: from the heart to the brain. J Neuroimmune Pharmacol 2: 313-318.

Zhao H. 2009. Ischemic postconditioning as a novel avenue to protect against brain injury after stroke. J Cereb Blood Flow Metab 29: 873-885.