Extended|Longevity

Scientific White Paper

Thyvolve Formula. Regenerates the thymus, increases T-cells, Includes phytotherapeutic extracts of: *Selaginella involvens*, Pinus sylvestris *(Pollen), Curcuma longa, Zingiber officinale, Elitaria cardamomum, and Cinnamomum verum*. Biological Actions, Molecular Mechanisms, and Their Effects.

Steven M. Schorr
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Thyvolve Formula, is designed for regenerating the Thymus Gland and reversing the age dependent process of Thymus involution. It is a synergistic herbal analog formulation of six (6) plant extracts including: ***Selaginella involvens*, *Pinus sylvestris (Pollen), Curcuma longa, Zingiber officinale, Elitaria cardamomum, and Cinnamomum verum***. **Thyvolve** is formulated to provide support for the regeneration of the Thymus.

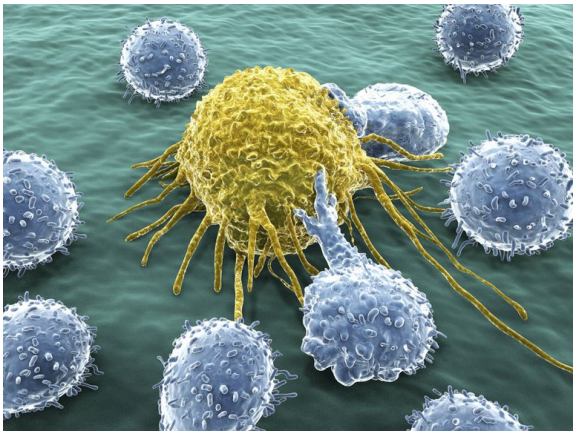
The Thymus Overview

The deterioration of the immune system with progressive aging is believed to contribute to morbidity and mortality in elderly humans due to the increased incidence of infection, autoimmunity, and cancer. The dysregulation of T-cell function is thought to play a critical part in these processes. One of the consequences of an aging immune system is the process termed "thymic involution", where the thymus undergoes a progressive reduction in size due to profound changes in its anatomy associated with loss of thymic epithelial cells and a decrease in thymopoiesis. This decline in the output of newly developed T cells results in diminished numbers of circulating naıve T cells and impaired cell-mediated immunity. This 'thymic menopause' includes the loss of thymic progenitors or epithelial cells, a diminished capacity to rearrange T-cell receptor genes and alterations in the production of growth factors and hormones. The thymus undergoes rapid degeneration following involution as part of the aging process. The thymus is capable of regenerating and restoring its function to a degree. Potential mechanisms for this endogenous thymic regeneration include keratinocyte growth factor (KGF) signaling and a more recently described pathway in which innate lymphoid cells produce interleukin-22 (IL-22). Thymic decline also occurs as an inevitable chronic process, in which the thymus gland undergoes involution with age. Thymic involution differs from aging in other organs that cannot be reversed. As the primary site of T cell development, the thymus plays a key role in the generation of a strong adaptive immune response, essential in the face of the potential threat from pathogens.

T Cells

T cells (T lymphocytes) derive their names from the organs in which they develop in the thymus. They arise in the bone marrow but migrate to the thymus gland to mature. The diverse responses of T cells are collectively called cell-mediated immune reactions. This is to distinguish them from antibody responses, which, of course, also depend on cells (B cells). T cells cannot recognize antigen alone, as for T cell receptors (TCRs),

they can recognize only antigens bound to cell-membrane proteins (MHC molecules). TCRs have different structures thus they bind to different molecular structures and have different genetic codes. Like antibody responses, T cell responses are exquisitely antigen-specific, and they are at least as important as antibodies in defending vertebrates against infection. Indeed, most adaptive immune responses, including antibody responses, require helper T cells for their initiation. Most importantly, unlike B cells, T cells can help eliminate pathogens that reside inside host cells.



Types of T Cells

T cells can be divided into four main classes:

1) Cytotoxic T cells directly kill infected cells by inducing them to undergo apoptosis, these cells like a "killer" or cytotoxin because they kill cells of interest that produce a particular antigen. The major surface marker of cytotoxic T cells is CD8, also known as killer T cells.

2) Helper T cells play an intermediate role in the immune response. They proliferate to activate B cells to make antibody responses and macrophages to destroy microorganisms that either invaded the macrophage were ingested by it. Helper T cells also help activate cytotoxic T cells to Helper T cells themselves but can only function when activated to become effector cells. The major surface marker of helper T cells is CD4.

3) Memory T cells consist of both CD4 and CD8 T cells that can rapidly acquire effector functions to kill infected cells and/or secrete inflammatory cytokines that inhibit replication of the pathogen. Together with memory B cells, lymphocytes that store specific antigen messages after antigen stimulation have lifespans of up to several decades. When they receive the same antigenic stimuli as they once again, they can proliferate as functional T cells against antigen or plasma cells that produce antibodies.

4) Regulatory / suppressor T cells often play an important role in maintaining tolerance and avoid excessive damage to the body's immune response. There are many classes of regulatory / suppressor T cells, including CD25 and CD4 T cells. They can inhibit T cells and B cells to regulate and control the immune response and maintain immune self-stability.

T Cell Differentiation

In the thymus, developing T cells, known as thymocytes, proliferate and differentiate along developmental pathways that generate functionally distinct subpopulations of mature T cells. Aside from being the main source of all T cells, it is where T cells diversity and then are shaped into an effective primary T cell repertoire by an extraordinary pair of selection processes. Cell differentiation is essential to create multiple subsets. Differentiation of naïve T cells into effector cells is required for optimal protection against different classes of the microbial pathogen and the development of immune memory. Differentiating cells undergo programmed alterations in their patterns of gene expression, which are regulated by structural changes in chromatin.

Differentiation is also directed by instructive and licensing signals from the environment, especially from antigen-presenting cells (APC). These cells gauge the class of the ingested microbe and generate signals that direct naïve T cells to differentiate into the subset that mobilizes the appropriate immune defense mechanisms. It is widely believed that cytokines are the major drivers of differentiation.

Diseases and Treatment of T Cell Differentiation

Helper T (Th) lymphocytes undergo two spatially and temporally distinct phases of differentiation. Following the first developmental phase, which occurs in the thymus, a second phase triggered by the initial encounter with antigen in the periphery leads to the development of effector T helper cell subsets displaying mutually exclusive patterns of cytokine gene expression. Clinically, Th1 patterns of cytokine production are associated with inflammation and autoimmune disease while Th2 patterns are characteristic of allergic responses and asthma. Understanding the complex process involved and the interaction between various cytokines, chemokines, signaling molecules, and pattern-recognition receptors (PRRs) in the immune pathways will provide valuable information for the development of novel therapeutic targets.

Active Herbal Ingredients

Selaginella involvens, the water extract of *S. involvens* showed a positive reaction to glycosides and possessed both thymus growth-stimulatory and antioxidant properties. It reverses involution of the thymus and exhibits remarkable antioxidant activity.

***Pinus sylvestris (Pollen*)**, Pine pollen is a potent source of androgenic substances composed of the bioidentical steroid hormone testosterone, along with lesser amounts of other steroids including androstenedione, dehydroepiandrosterone (DHEA) and androsterone. Pine pollen also contains estrogens including estrone, estriol, and estradiol, as well as progesterone.

Curcuma longa, or Turmeric root, and its curcuminoid constituents have demonstrated properties consistent with decreases in inflammatory stress signaling and increases in protective signaling. Curcumin is known to have anti-aging, anti-oxidant, anti-inflammatory, anti-arthritic, and anti-cancer effects and increases BDNF while having a positive effect on Alzheimer's disease and depression. It is also anti-rheumatic, and anti-microbial.

Zingiber officinale, Ginger, is an excellent source of phenolic compounds including gingerols, shogaols, paradols, and gingerdiones. Literature has extensively demonstrated that 6-gingerol (6G), 8-gingerol (8G), 10-gingerol (10G) and 6-shogaol (6S) collectively referred to as ginger phenolics (GPs,), are the most abundant bioactive constituents of whole ginger extract, and exhibit anti-proliferative, anti-inflammatory, anti-oxidative and anti-tumor properties.

Elitaria cardamomum, is one of the highest sources of plant-based zinc. Extracts of cardamom may be effective against a variety of bacterial strains that contribute to fungal infections.

Cinnamomum verum, Cinnamon health benefits are attributed to its content of a few specific types of antioxidants, including polyphenols, phenolic acid and flavonoids. These compounds work to fight oxidative stress in the body and aid in the prevention of chronic disease. the effects of cinnamon on life span implicated major longevity pathways. These include the DAF-16 transcription factor in the insulin signaling pathway, which promotes expression of stress resistance and the longevity genes. Cinnamon activates the insulin signaling pathway, anti-oxidative pathway and serotonin signaling for its lifespan prolonging effect.

Extended Longevity

Arch. Immunol. Ther. Exp. (2017) 65:401–420
DOI 10.1007/s00005-017-0462-x

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REVIEW

Acute Thymic Involution and Mechanisms for Recovery

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Received: 13 June 2016 / Accepted: 12 March 2017 / Published online: 22 March 2017

Abstract Acute thymic involution (ATI) is usually regarded as a virulence trait. It is caused by several infectious agents (bacteria, viruses, parasites, fungi) and other factors, including stress, pregnancy, malnutrition and chemotherapy. However, the complex mechanisms that operate during ATI differ substantially from each other depending on the causative agent. For instance, a transient reduction in the size and weight of the thymus and depletion of populations of T cell subsets are hallmarks of ATI in many cases, whereas severe disruption of the anatomical structure of the organ is also associated with some factors, including fungal, parasitic and viral infections. However, growing evidence shows that ATI may be therapeutically halted or reversed. In this review, we highlight the current progress in this field with respect to numerous pathological factors and discuss the possible mechanisms. Moreover, these new observations also show that ATI can be mechanistically reversed.

Keywords Acute · Thymus · Atrophy · Thymic involution · Mechanistic recovery

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Introduction

The thymus is a unique lobulated lymphoid tissue that develops as a gland (Bódi et al. 2015). It is considered a primary immune organ in jawed vertebrates, in which it plays important roles in the selection, proliferation, development and differentiation of T cells and provides protection against infections by various pathogens (Bajoghli and Guo 2011; Gameiro et al. 2010; Liu et al. 2014). Despite its significance in immunity (Miller 2002; Samara et al. 2016), the shrinkage of thymus that occurs with aging results in a decrease in tissue mass along with architectural alterations. In general, almost all vertebrates that possess a thymus experience this ancient and conserved evolutionary process, termed age-related thymic involution (Shanley et al. 2009), although the thyme of some shark species are reported not to undergo involution (Zakharova 2009). However, under certain physiological and pathological situations, the thymus may abruptly undergo a transient regression, known as acute thymic involution (ATI) (Shanley et al. 2009). In age-related thymic atrophy, the maximum decline in the thymic weight occurs just before the start of the mid-phase of life (Aspinall and Andrew 2000), i.e., at approximately 30–40 years of age in humans (Bertho et al. 1997) and 9–12 months of age in mice (Aspinall 1997). After this point, it involutes slowly during further aging (Aspinall and Andrew 2000). In contrast, ATI is usually seen during gestational (Ekin et al. 2016; Jacques et al. 2014, 2015) and neonatal life (Eriksen et al. 2014; Nickels et al. 2015; Toti et al. 2000) in humans. However, ATI in experimental animal models has been reported to occur not only during gestational periods (Kunzmann et al. 2010; Kuypers et al. 2012) and neonatal life (Falkenberg et al. 2014; Zhou et al. 2016), but also in adult life stages (Lee et al. 2011; Park et al. 2007). However, the basic difference

Published in final edited form as:
Curr Opin Pharmacol. 2010 August ; 10(4): 443–453. doi:10.1016/j.coph.2010.04.008.

Emerging strategies to boost thymic function

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Abstract

The thymus constitutes the primary lymphoid organ for the generation of T cells. Its function is particularly susceptible to various negative influences ranging from age-related involution to atrophy as a consequence of malnutrition, infection or harmful iatrogenic influences such as chemotherapy and radiation. The loss of regular thymus function significantly increases the risk for infections and cancer because of a restricted capacity for immune surveillance. In recent years, thymus-stimulatory, -regenerative and -protective strategies have been developed to enhance and repair thymus function in the elderly and in individuals undergoing hematopoietic stem cell transplantation. These strategies include the use of sex steroid ablation, the administration of growth and differentiation factors, the inhibition of p53, and the transfer of T cell progenitors to alleviate the effects of thymus dysfunction and consequent T cell deficiency.

Keywords

thymus

Introduction

The principal function of the immune system is to resist infectious agents and to eliminate malignantly transformed cells. This challenging task is effected by two closely interacting defense mechanisms known as the innate and the adaptive immune systems. The innate arm provides an immediate but generally non-specific protection that is activated by pattern recognition receptors identifying structural components well conserved among different pathogens. In contrast, the adaptive arm of the immune response is characterized by antigen-specificity and immunological memory and is achieved by two interdependent systems: Antibody-producing B-lymphocytes and the cellular defense of T lymphocytes.

Both lymphoid cell types arise from pluripotent hematopoietic stem cells (HSC) resident in the bone marrow. In contrast to B cell development that largely takes place in the bone marrow, T cell maturation requires the specialized microenvironment of the thymus as a site for lineage commitment and differentiation. The thymus is located in the upper anterior mediastinum, is structured into an outer cortex and an inner medulla and continuously generates T cells that are exported to the periphery. Intrathymic T-cell development constitutes an intricate process of cellular cross-talk where an array of interacting stromal

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Insights into thymic aging and regeneration

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Acknowledgements
We thank Drs Vishwa Deep-Dixit and Valeria Coelho and Ms. Ana Lustig for their hard work on some of the projects discussed in this review and assistance with this manuscript. We also thank Ms. Angela Feehley for her editorial assistance.

Immunological Reviews 2005
Vol. 205: 72–93
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Immunological Reviews
0105-2896

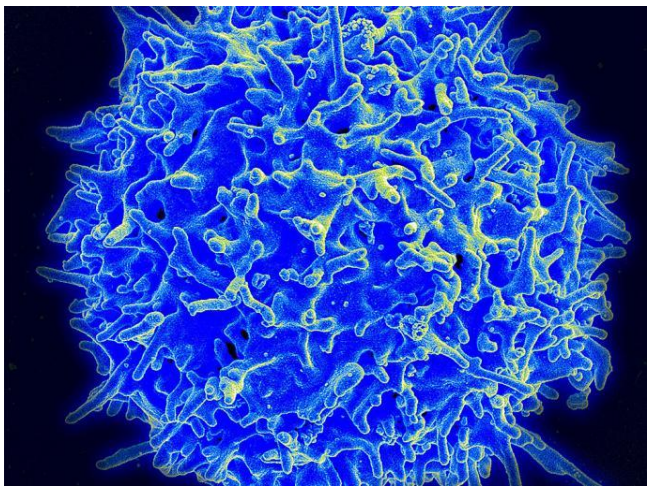
Summary: The deterioration of the immune system with progressive aging is believed to contribute to morbidity and mortality in elderly humans due to the increased incidence of infection, autoimmunity, and cancer. Dysregulation of T-cell function is thought to play a critical part in these processes. One of the consequences of an aging immune system is the process termed thymic involution, where the thymus undergoes a progressive reduction in size due to profound changes in its anatomy associated with loss of thymic epithelial cells and a decrease in thymopoiesis. This decline in the output of newly developed T cells results in diminished numbers of circulating naïve T cells and impaired cell-mediated immunity. A number of theories have been forwarded to explain this 'thymic menopause' including the possible loss of thymic progenitors or epithelial cells, a diminished capacity to rearrange T-cell receptor genes and alterations in the production of growth factors and hormones. Although to date no interventions fully restore thymic function in the aging host, systemic administration of various cytokines and hormones or bone marrow transplantation have resulted in increased thymic activity and T-cell output with age. In this review, we shall examine the current literature on thymic involution and discuss several interventional strategies currently being explored to restore thymic function in elderly subjects.

Introduction

Age-associated alterations in immune function both in humans and in animals are important to the health of aging individuals. Older humans are more susceptible to microbial infections of the urinary and respiratory tracts, soft tissue, skin, and the abdominal region (1, 2). Moreover, elderly subjects demonstrate an increased incidence of infectious endocarditis, meningitis, tuberculosis, and herpes zoster, and the mortality rates for these diseases in older patients are often two to three times higher than in younger people with the same disease (3–6). Additionally, elderly subjects also demonstrate an increased prevalence of specific cancers and certain autoimmune diseases (6–9). The increased prevalence of these conditions and the higher morbidity and mortality from infections strongly suggest functional defects from a deteriorating immune system with advancing age (10–16).

Restoring Thymus Function for Enhanced Immune Response

By **NutritonReview.org** - September 12, 2013



The key to a healthy, functioning immune system rests with the thymus gland, a small organ lying just beneath the breastbone. The primary role of the thymus is to assist in the proliferation and differentiation of mature T-lymphocytes – cells that attack and kill viruses and bacteria. T-cells (Fig. 1) emerge from the bone marrow in an incomplete state. In order to function properly, immature T-cells migrate to the thymus gland where they are programmed into mature T-cells that orchestrate the immune response to attack and destroy invading viruses and cancer cells.

In our early twenties we have an abundance of well-trained, functioning T-cells that regulate the immune system and help the body fight off pathogens and disease. After about age 20, the thymus begins to shrink (atrophy) as dying thymic cells are progressively replaced by fat and connective tissue.

By about age 40, output of thymic hormones has dropped significantly and T-cells have begun to lose their effectiveness. It is this gradual loss of functioning T-cells that is believed to be responsible for many of the age-related changes in the immune system that gradually rob the body of its ability to fight off infectious diseases, autoimmune disorders and cancer.

Antiaging Effects of Vital Cell in Rabbits

To evaluate the effects of *Vital Cell* on organ health, researchers conducted a two-year trial with two identical groups of rabbits. One group was treated with Vital Cell daily, and the second, untreated group served as a control. At the end of the study, the researchers compared the organs of both test groups of treated and untreated elderly rabbits to those of young, healthy juvenile rabbits. When examining the treated rabbits the researchers noted that thymus glands of the old animals receiving Vital Cell retained the structure and functionality of glands normally seen only in young, health rabbits.

Conversely, the thymus glands of the old, untreated control rabbits were severely atrophied, weighing less than a third of their normal weight, and consisting primarily of inactive fat and

Published in final edited form as:
Immunol Rev. 2016 May ; 271(1): 56–71. doi:10.1111/imr.12418.

Thymus: The Next (Re)Generation

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Summary

As the primary site of T cell development, the thymus plays a key role in the generation of a strong yet self-tolerant adaptive immune response, essential in the face of the potential threat from pathogens or neoplasia. As the importance of the role of the thymus has grown, so too has the understanding that it is extremely sensitive to both acute and chronic injury. The thymus undergoes rapid degeneration following a range of toxic insults, and also involutes as part of the aging process, albeit at a faster rate than many other tissues. The thymus is, however, capable of regenerating, restoring its function to a degree. Potential mechanisms for this endogenous thymic regeneration include keratinocyte growth factor (KGF) signaling, and a more recently described pathway in which innate lymphoid cells produce interleukin-22 (IL-22) in response to loss of double positive thymocytes and upregulation of IL-23 by dendritic cells. Endogenous repair is unable to fully restore the thymus, particularly in the aged population, and this paves the way towards the need for exogenous strategies to help regenerate or even replace thymic function. Therapies currently in clinical trials include KGF, use of the cytokines IL-7 and IL-22, and hormonal modulation including growth hormone administration and sex steroid inhibition. Further novel strategies are emerging in the pre-clinical setting, including the use of precursor T cells and thymus bioengineering. The use of such strategies offers hope that for many patients, the next regeneration of their thymus is a step closer.

Keywords

Thymus damage; Aging; Tissue Regeneration

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Conflict of interest: The authors are not aware of any affiliations, memberships, funding, or financial holdings that might be perceived as affecting the objectivity of this review. Patent applications have been filed on the therapeutic use of IL-22 (US 61/487,517; US 61/901,151) with J.A.D, A.M.H, and M.R.M vdB listed as inventors.

PubMed thymus growth-stimulatory component from Selaginella involvens

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Immunopharmacol Immunotoxicol. 2011 Jun;33(2):351-9. doi: 10.3109/08923973.2010.518617.

Protection of immunocompromised mice from fungal infection with a thymus growth-stimulatory component from Selaginella involvens, a fern.

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Abstract

CONTEXT: Recent studies have shown that the water extract of **Selaginella involvens** (Sw.) Spring, a wild fern, exhibits **thymus growth-stimulatory** activity in adult mice (reversal of involution of **thymus**) and remarkable anti-lipid peroxidation activity. Follow-up studies were carried out in the present study.

MATERIALS AND METHODS: Activity-guided isolation of the active **component** (AC) was carried out. The effect of AC on immune function was studied using fungal (Aspergillus fumigatus) challenge in cortisone-treated mice. The in vitro antifungal activity of AC was assayed using disc diffusion assay. In vitro and in vivo effect of AC on DNA synthesis in **thymus** was studied using (3)H-thymidine incorporation. In in vitro anti-lipid peroxidation, hydroxyl radical scavenging and inhibition of superoxide production were assayed.

RESULTS: The active principle/**component** (AC) was isolated in a chromatographically pure form from the water extract of S. **involvens**. AC showed positive reaction to glycosides. AC possessed both **thymus growth-stimulatory** and antioxidant properties. It protected cortisone-treated mice from A. fumigatus challenge. It did not exhibit in vitro antifungal activity. Increased (3)H-thymidine incorporation was observed in the reticuloepithelium of **thymus** obtained from AC-treated mice. However, in vitro AC treatment to **thymus** for 5 h did not result in an increase in (3)H-thymidine incorporation.

DISCUSSION AND CONCLUSION: AC (named as Selagin), from S. **involvens**, could reverse

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SCIENTIFIC REPORTS

OPEN

Pharmacokinetic-pharmacodynamic correlations in the development of ginger extract as an anticancer agent

Received: 29 June 2017
Accepted: 29 January 2018
Published online: 14 February 2018

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Anticancer efficacy of ginger phenolics (GPs) has been demonstrated in various *in vitro* assays and xenograft mouse models. However, only sub-therapeutic plasma concentrations of GPs were detected in human and mouse pharmacokinetic (PK) studies. Intriguingly, a significant portion of GPs occurred as phase II metabolites (mainly glucuronide conjugates) in plasma. To evaluate the disposition of GPs and understand the real players responsible for efficacy, we performed a PK and tissue distribution study in mice. Plasma exposure of GPs was similar on day 1 and 7, suggesting no induction or inhibition of clearance pathways. Both free and conjugated GPs accumulated in all tissues including tumors. While non-cytotoxicity of 6-ginerol glucuronide precluded the role of conjugated GPs in cell death, the free forms were cytotoxic against prostate cancer cells. The efficacy of ginger was best explained by the reconversion of conjugated GPs to free forms by β-glucuronidase, which is over-expressed in the tumor tissue. This previously unrecognized two-step process suggests an instantaneous conversion of ingested free GPs into conjugated forms, followed by their subsequent absorption into systemic circulation and reconversion into free forms. This proposed model uncovers the mechanistic underpinnings of ginger's anticancer activity despite sub-therapeutic levels of free GPs in the plasma.

Recently, there has been a resurgence of whole food-based therapeutics categorized as Natural Medicines (NMs) in prevention and treatment of chronic diseases like cancer[1,2]. However, their clinical development has suffered perhaps due to sub-therapeutic concentrations of constituent phytochemicals in the plasma. Ginger is an example. An almost instantaneous conversion of the ingested free ginger phenolics (GPs) into their pharmacologically-inactive phase II metabolites (glucuronides, sulfates, etc.) in the plasma has perpetuated a diminished interest in their development. However, this highlights a knowledge gap as to how constituent phytochemicals contribute physiologically to the observed pharmacological effects of NMs. Undoubtedly, NMs are a complex concoction of chemically-diverse phytochemicals containing one to several bioactive compounds, which mostly are plant secondary metabolites. Recently, a unique approach called '*reverse pharmacokinetics*' has helped to draw possible reasons underlying the discordance between pharmacokinetics (PK) and pharmacodynamics (PD) of NMs[3]. In this study, we examined how the free (active) and conjugated (inactive) species interconvert on the physiological time scale to ultimately pinpoint the bioactive species responsible for the PD effect. For almost a decade now, our laboratory has been studying the pharmacology of ginger root extract. Thus, ginger extract was our obvious choice as the probe NM to investigate the pharmacokinetic-pharmacodynamic (PK-PD) relationships between free and conjugated forms of GPs to precisely define the role of Phase II metabolites in the anticancer activity of ginger extract.

Ginger (*Zingiber officinale*), a widely consumed spice worldwide, is an excellent source of phenolic compounds including gingerols, shogaols, paradols, gingerdiones etc.[4–6]. Literature has extensively demonstrated that 6-gingerol (6G), 8-gingerol (8G), 10-gingerol (10G) and 6-shogaol (6S) collectively referred to as ginger phenolics (GPs, Supplementary Figure 1), are the most abundant bioactive constituents of whole ginger extract, and exhibit anti-proliferative, anti-inflammatory, anti-oxidative and anti-tumor properties[4,7–12]. We have published the tumor growth-inhibitory efficacy of ginger extract (GE) in prostate cancer models as well as reported

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