

REVIEW ARTICLE

Helminth-induced apoptosis: a silent strategy for immunosuppression

AMIN ZAKERI*

Department of Pathobiology, Faculty of Veterinary Medicine, Ferdowsi University of Mashhad, Mashhad, Iran

(Received 23 March 2017; revised 1 May 2017; accepted 9 May 2017; first published online 29 June 2017)

SUMMARY

During microbial infections, both innate and adaptive immunity are activated. Viruses and bacteria usually induce an acute inflammation in the first setting of infection, which helps the eliciting an effective immune response. In contrast, macro-parasites such as helminths are a highly successful group of invaders known to be capable of maintaining a chronic infestation with the minimum instigation. Undoubtedly, generating such an immunoregulatory environment requires the exploitation of various immunosuppressive mechanisms to debilitate host immunity supporting their survival and replication. Several mechanisms have been recognized whereby helminths prolong their infections including an increase of immunoregulatory cells, inhibition of Th1 or Th2 responses, targeting pattern recognition receptors (PRRs) and lowering the immune cells quantity via induction of apoptosis. Apoptosis is a programmed intracellular process involving a series of consecutive downstream signalling event evolved to cell death. It plays a pivotal role in several immunological reactions in particular deletion of autoreactive immune cells. Helminth-triggered apoptosis in immune cells exhausts host immunity, which paves the way for generating a permissive environment and chronic infection. This review provides a compilation of recent investigations discussing the apoptotic mechanisms exploited by different worms and the immunological consequences of immune cell death. Finally, the anti-cancer effects of some worm-derived molecules due to their apoptotic effects are discussed, highlighting as potentially druggable candidates to combat cancer.

Key words: Apoptosis, helminth, host–parasite interaction, immunosuppression.

INTRODUCTION

Apoptosis is an important process, which physiologically occurs in throughout of mammalian life. It has been indicated as one type of programmed cell death, which enables organisms to eliminate injured cells from the body (Devitt and Marshall, 2011). Also, it plays an essential role in tissue homeostasis and fundamental processes of the immune system such as central and peripheral tolerance, setting up immunological memory, and negative selection (Opferman, 2008). During the process of apoptosis, several intracellular signalling pathways are triggered, which results in recruitment and activation of a series of proteases known as caspases (Devitt and Marshall, 2011).

Different pathogens including viruses, bacteria and protozoa have developed strategies to induce apoptosis in both host immune and non-immune cells (Luder *et al.* 2001; Herold *et al.* 2012). *Listeria monocytogenes*, for example, is able to trigger apoptosis in host lymphocytes as an important mechanism for survival (Carrero and Unanue, 2006).

* Corresponding author. Department of Pathobiology, Faculty of Veterinary Medicine, Ferdowsi University of Mashhad, Mashhad, Iran. E-mail: Aminzakeri7@gmail.com, Amin.zakeri@mail.um.ac.ir

Intracellular pathogens, such as *Toxoplasma gondii* and *Leishmania* can also heighten their infectivity by deriving apoptosis-related signalling (Bienvenu *et al.* 2010).

Worms are adept at bypassing host immunity, as during many helminth infections the host immune responses are modulated and quietly suppressed. Notably, a number of investigations imply that induction of apoptosis by helminths is likely to play an important role in dampening host immunity (Chen *et al.* 2008; Gazzinelli-Guimaraes *et al.* 2013). Promotion of host cell death, especially in immune cells facilitates the parasite proliferation, as well as increases the longevity of helminths within the host through lowering the quantity of immune cells (Chow *et al.* 2000).

Helminths and their products have been found to trigger apoptosis pathways and anergy in host immune cells including T lymphocytes (Chow *et al.* 2000; O'Connor *et al.* 2003; Smith *et al.* 2004; Chen *et al.* 2008), antigen presenting cells (APCs), natural killer cells (NK cells) and eosinophils (Moreau and Chauvin, 2010; Babu and Nutman, 2012). Besides immune cells, non-immune cells such as intestinal epithelial cells (Cliffe *et al.* 2007) are also targeted by helminths and their products for apoptosis.

Two general pathways are involved in apoptosis process including the death receptor pathway and mitochondrial pathway (Devitt and Marshall, 2011). Stimulation of these pathways by helminths indicates that these macroparasites during their complex interactions with host immunity have evolved complex mechanisms to promote their lifespan within the host. Some helminths migrate throughout the host body and their large size can induce stress signals and local inflammation in afflicted tissues.

It seems that their main purpose of killing immune cells is likely dampening inflammatory responses raised against them. Thus, it is not surprising to hypothesize induction of apoptosis is a telling mechanism to suppress inflammation and pave the way for immunoevasion.

Undoubtedly, unravelling the principal pathways through which helminths manipulate viability of host cells will represent new insights into their immunoregulatory functions, leading us to the development of new approaches for fighting helminth infections. On the other hand, recognition of bioactive molecules whereby helminths induce apoptosis may offer potential drugs to combat diseases such as cancers in which aberrant cell division and long lifespan are the major problems (Vasilev *et al.* 2015).

This review aims to highlight the recent findings concerning the interactions between helminths and host cells resulting in apoptosis as a powerful and salient mechanism for immunosuppression.

APOPTOSIS

Apoptosis is regarded as a complex process recruiting different intracellular molecules to drive involved cells toward programmed death. Development of biochemical interaction between upstream and a key family of downstream cysteine proteases known as caspases enables an organism to eliminate afflicted and old cells via this irreversible process. Apoptosis plays a pivotal role not only in development and homeostasis of the immune system, but also in generation and maintenance of immunologic tolerance to antigens (Kushwah and Hu, 2010).

Generally, the main routes triggering apoptosis are categorized into two divergent pathways called the intrinsic (mitochondrial) and the extrinsic (death receptor) pathways. The major activators of the intrinsic pathway are factors making DNA damage and endoplasmic reticulum stress (ERS). But, the extrinsic pathway is triggered by a ligand–receptor interaction between tumour necrosis factor (TNF) family including TNF α , TRAIL (TNF-related apoptosis-inducing ligand) and Fas ligand and the surface receptors known as TNF receptor (TNFR) superfamily such as TNFR1 and 2, death receptor 4/5 and Fas. Both pathways eventually lead to activation of caspases (Bai and Wang, 2014).

The intrinsic pathway is triggered upon any stimulation inducing mitochondrial outer membrane permeability (MOMP) due to pore formation. In fact, MOMP is the central player, which mediates subsequent events including the release of cytochrome c and second mitochondria-derived activator of caspases (SMAC) as proapoptotic proteins to the cytoplasm. Cytochrome c actively contributes to the formation of the caspase-9-activating complex known as the apoptosome via binding to the adaptor protein apoptotic protease-activating factor 1 (APAF-1). Upon constitution of apoptosome, caspases 3 and 7 are activated, which ultimately results in DNA fragmentation and emergence of other apoptosis-related signs. In the extrinsic pathway, engagement of death receptor leads to the formation of a multiprotein complex called death-inducing signal complex that activates caspase 8. Subsequently, caspase 8 is able to cleave and activate caspase 3 and 7, the executor caspases in programmed cell death (Fernald and Kurokawa, 2013).

In this review, the association of helminths and immune system apoptosis is detailed below according to the parasite species, taking in turn trematodes (*Fasciola* and *Schistosoma*), nematodes (*Filarial* and *Trichinella*) and the cestodes (*Taenia* and *Echinococcus*). A summary of the pathways and mechanisms through which helminths target mainly immune cells to undergo apoptosis has been referred in Table 1.

TREMATODES

Fasciola hepatica

Fasciola hepatica involves a wide range of animals, including ruminants, rodents and humans (Robinson and Dalton, 2009). This worm infects through the duodenum and intestinal wall, enters the peritoneal cavity where it penetrates in the liver capsule and resides there (Robinson and Dalton, 2009; Cwiklinski *et al.* 2016). In spite of insulting various tissues and eliciting immune responses, *F. hepatica* can survive in the host for a long time. Induction of host cell apoptosis by *F. hepatica* and its products has been well recognized as an efficacious mechanism to suppress host immunity (Serradell *et al.* 2007, 2009; Guasconi *et al.* 2012; Escamilla *et al.* 2016). In this regard, excretory-secretory products (ESP) from *F. hepatica* are able to induce eosinophil apoptosis via triggering a series of mitochondrial-dependent pathways. The most upstream pathway activated by ESP that results in eosinophil apoptosis is tyrosine kinases (Tyr K) pathway, as inhibition of this pathway abrogates apoptotic effects of ESP (Serradell *et al.* 2007).

Serradell *et al.* (2009) studied the main mechanism through which both live worm and its ESP mediate eosinophil apoptosis. In this investigation, it was revealed that ESP through increasing the production

Table 1. Major mechanisms and involved pathways during induction of host cell apoptosis by helminths and their products

Worm	Live worm/component	Targeted cells to induce apoptosis	Major findings/mechanism	References
<i>Fasciola hepatica</i>	ESP	Eosinophils	Tyrosine kinases (TyrK) and caspases	Serradell <i>et al.</i> (2007)
	Live worm and ESP	Eosinophils	Induction of ROS and H ₂ O ₂ causes mitochondrial-membrane depolarization and release of cytochrome c	Serradell <i>et al.</i> (2009)
<i>Schistosoma mansoni</i>	Live worm	Eosinophils in sheep liver	Caspase3-dependent	Escamilla <i>et al.</i> (2016)
	Metacercariae and ESP	Macrophages	Unknown	Guasconi <i>et al.</i> (2012)
	SEA	Hepatic stellate cells (HSC)	Activation of caspase 3, 8, and stimulation of (TRAIL/DR5)	Duan <i>et al.</i> (2014)
	SEA	Immortalized human hepatic stellate cell line (LX-2)	Increase of p53 and DR5 expression and decrease of Akt expression	Wang <i>et al.</i> (2014)
<i>Trichinella spiralis</i>	Cercariae	CD4 ⁺ T cells	IL-10-dependent mechanism	Prendergast <i>et al.</i> (2015)
	Recombinant protein (rSj16)	Murine myeloid leukemia WEHI-3B JCS cells	Suppressing G0/ G1 phase, promotes apoptosis by targeting mitochondrial membrane potential, increase of caspase 3, 6, and 9 activity, up-regulate pro-apoptotic Bax expression and down-regulate anti-apoptotic Bcl-2 expression	Yang <i>et al.</i> (2013)
	SEA and Live worm	CD4 ⁺ T cells	Through Fas Ligand-expressing B-1a lymphocytes	Lundy <i>et al.</i> (2001), Lundy and Boros (2002)
	Smaf	CD4 ⁺ T cells and CD8 ⁺ T cells	Increasing the expression of Fas, FasL, and the Fas-associated death domain in skin-related T cells	Chen <i>et al.</i> (2002)
<i>Trichinella spiralis</i>	Live worm	Intestine cells	Cleavage of caspase 12 by ERS-related molecules and phosphorylation of JNK	Yu <i>et al.</i> (2014)
	(ES L1) muscle larvae	B16 melanoma cells	Activation of the outer caspase-dependent apoptotic pathway and death receptor	Vasilev <i>et al.</i> (2015)
	Live worm	Tumour-bearing mice	Inhibition of the development and metastasis of melanoma tumour by modulation of cytokine profile (apoptosis mechanism is unknown)	Kang <i>et al.</i> (2013)
<i>Trichinella spiralis</i>	Crude extract	Different cancerous cell lines and Tumour-bearing mice	Reduction of cell proliferation and tumour growth (apoptosis mechanism is unknown)	Wang <i>et al.</i> (2009), Wang <i>et al.</i> (2013)
	Secretory antigens (BpA)	Monocytes	Caspase3 and TLR4-dependent	Das Mohapatra <i>et al.</i> (2014)
<i>Brugia pahangi</i>	L3	NK cells	Caspase-dependent pathway	Babu <i>et al.</i> (2007)
	Live worm	CD4 ⁺ T Cells	Up-regulation of FasL (death ligand) in B-1 cells	Mishra <i>et al.</i> (2017)
<i>Onchocerca volvulus</i>	OvALT-2, OvNLT-1	Ovalbumin-specific CD4 ⁺ DO11.10, OT-II T cells, and CD8 ⁺ OT-II cells	Suppression of DNA synthesis, cell division, and cytokine production	Hartmann <i>et al.</i> (2013)
	Live worm	Leucocytes of dogs	Unknown but correlation between apoptosis and oxidative status	Dimiri <i>et al.</i> (2012)
<i>Dirofilaria immitis</i>	Live worm	CD4 ⁺ T lymphocytes	Unknown	Tato <i>et al.</i> (2004), Solano <i>et al.</i> (2006), Sikasunge <i>et al.</i> (2008)
	Metacystode			

Table 1. (Cont.)

Worm	Live worm/component	Targeted cells to induce apoptosis	Major findings/mechanism	References
<i>Taenia solium</i>	Annexin B1 (a metacercaria-derived compound)	Eosinophils	Activation of caspase 3 and cytochrome c through induction of Ca ²⁺ influx (mitochondrial pathway)	Yan <i>et al.</i> (2008)
<i>Taenia crassiceps</i>	Metacercodes	B and T lymphocytes including CD4 ⁺ , CD8 ⁺ , and CD19 ⁺ and eosinophils	Increase of caspase-3 activity	Zepeda <i>et al.</i> (2010)
<i>Echinococcus multilocularis</i>	MF Cysticercal antigens	Spleen CD4 T lymphocytes CD4 ⁺ and CD19 ⁺ splenocytes	Unknown Unknown	Zepeda <i>et al.</i> (2016) Lopez-Briones <i>et al.</i> (2003)
<i>Paragonimus westermani</i>	larval-derived vesicles	DCs	Unknown	Nono <i>et al.</i> (2012)
<i>Nippostrongylus brasiliensis</i>	Live worm ESP of metacercariae ESP	Hepatocytes Eosinophils Intestinal epithelial cell line IEC-6	Up-regulation of genes supporting hepatocyte apoptosis such as p53, p21, Gadd45c and cleaved-caspase 3 ESP induce phosphatidylserine (PS) externalization on the outer surface of eosinophils along with caspase3 activation Up-regulation of Fas (CD95) and activation of caspase 3, but the main mechanism is unknown	Zhang <i>et al.</i> (2012) Min <i>et al.</i> (2004) Kuroda <i>et al.</i> (2002)

SEA, soluble antigens of eggs; ERS, endoplasmic reticulum stress, OvALT-2, *Onchocerca volvulus* abundant larval transcript-2; OvNLT-1, *Onchocerca volvulus* novel larval transcript-1; Smaf, *S. mansoni*-derived apoptosis-inducing factor; MF, metacercode factor, L3, live infective-stage larvae.

of reactive oxygen species (ROS), in particular, H₂O₂ causes mitochondrial-membrane depolarization evolving to release of cytochrome c and consequently activation of caspase cascade (Serradell *et al.* 2009). Intraperitoneal administration of catalase as H₂O₂ scavenger enzyme to rats infected with *F. hepatica* inhibited the eosinophil apoptosis, as well as exposing ESP-treated eosinophil to catalase suppressed apoptosis, suggesting the critical role of H₂O₂ in ESP and *F. hepatica*-mediated apoptosis. ESP also was found to stimulate eosinophil apoptosis through caspase-dependent manner. The critical caspases involved in ESP-induced apoptosis are caspase3, 8 and 9 (Serradell *et al.* 2009). To confirm the involvement of caspase pathway in ESP-induced apoptosis, Z-VAD-fmk as a caspase inhibitor was used, which could forestall apoptosis of eosinophil. Further experiments by Serradell *et al.* (2009) showed that carbohydrate components present in ESP crude antigens are responsible for induction of eosinophil apoptosis.

Evaluation of *F. hepatica*-induced apoptosis in the liver of sheep, as the most important natural host of this worm, has recently provided interesting findings. Migration of the worm in the liver of sheep orally infected with metacercariae can result in apoptosis of liver eosinophils. Based on immunohistochemistry and transmission electron microscopy findings (Escamilla *et al.* 2016), the presence of abundant caspase 3⁺ and nuclear-fragmented eosinophils at the necrotic sites of liver and bile ducts shows that *F. hepatica* can efficiently induce apoptosis in both migratory and biliary stages (Escamilla *et al.* 2016).

Besides eosinophils, macrophages also have been shown to be killed by ESP through apoptosis. Guasconi *et al.* (2012) during an *in vitro* study evaluated the effects of FhESP on peritoneal macrophages and cells obtained from mice infected with metacercaria. In this study, propidium iodide (PI) staining was used to confirm the presence of hypodiploid nuclei in cells exposed to FhESP (Guasconi *et al.* 2012). High levels of hypodiploid cells were detected in macrophages treated with FhESP and cells derived from mice infected with metacercaria. Induction of apoptosis was further confirmed when the results of annexin-V assay showed a significant increase of annexin-V positive cells (Guasconi *et al.* 2012).

It seems that induction of apoptosis in eosinophils and macrophages as essential immune cells fighting against helminths is a telling mechanism through which *F. hepatica* overcomes host immune responses. However, more studies are required to clarify whether other immune cells are primed to undergo apoptosis by this worm.

Schistosoma spp.

Schistosomiasis involves a wide range of animals in worldwide due to infection with *Schistosoma* species

including *Schistosomiasis mansoni*, *Schistosomiasis japonicum*, and *Schistosomiasis haematobium* (Barsoum *et al.* 2013). Cercariae are schistosome larvae, which live in freshwater. They penetrate into the mammalian host through the skin where transform into schistosomula, then entry into blood vessels and migrate toward lungs and liver (Barsoum *et al.* 2013). During this long survey, they masterfully exploit intriguing mechanisms to evade host immunity including altering membrane antigens, antibody cleavage and apoptosis of host cells (Carneiro-Santos *et al.* 2000; Burke *et al.* 2009).

Liver damage is one of the most prevalent clinical manifestations of schistosomiasis, which occurs as a result of immune responses against trapped eggs in this organ. The presence of too narrow vessels in the liver known as sinusoids avoids egg transition and entraps them, which eventually leads to pathologic outcomes such as granuloma and fibrosis (Pearce and MacDonald, 2002). It has been reported that the eggs of *S. mansoni* contain soluble egg antigens (SEA) diminishing liver fibrosis via apoptosis of hepatic stellate cells (HSC), which have a pivotal role in the progress of liver fibrosis. It seems that SEA stimulates the extrinsic pathway of apoptosis in HSCs. Activation of caspase 3 and 8 along with stimulation of TNF-related apoptosis-inducing ligand/death receptor 5 (TRAIL/DR5) are the major mechanisms by which the SEA induces apoptosis (Duan *et al.* 2014). Further *in vitro* experiments showed that SEA through the increase of p53 and DR5 expression and a decrease of protein kinase B (PKB), known as Akt, expression promotes apoptosis in the immortalized human hepatic stellate cell line (LX-2) (Wang *et al.* 2014). However, the nature of such an anti-fibrotic function of *S. mansoni* in the liver is really unknown.

As mentioned earlier, *S. mansoni* larvae are able to pierce the skin to accede to the blood vessels of the definitive host. Undoubtedly, this offense will result in local inflammation and activation of APCs and consequently stimulation of CD4⁺ T lymphocytes. Recently, Prendergast *et al.* (2015) demonstrated that re-exposure of mice to the high number of *S. mansoni* cercariae (600 cercariae) induces hyporesponsiveness and apoptosis in CD4⁺ T cells via an IL-10-dependent mechanism, whereas exposure of mice to a single dose of infection (150 cercariae) did not affect CD4⁺ T cells. The apoptotic effects of *S. mansoni* on CD4⁺ T cells had already been proved by Lundy and colleagues. They indicated that Fas ligand-expressing B-1a lymphocytes play an essential role in triggering apoptosis in CD4⁺ T cells during both schistosomal infection and exposure to SEA (Lundy *et al.* 2001; Lundy and Boros, 2002). Skin-stage schistosomula of *S. mansoni* have also been found to be equipped to immunosuppressive molecules enabling them to frustrate early cellular responses and evade recognition in the

host skin (Chen *et al.* 2002). In support of this, molecular dissection has revealed that skin-stage schistosomula is able to release a potent pro-apoptotic molecule that significantly triggers apoptosis in skin-associated CD4⁺ and CD8⁺ T cells via stimulating Fas/FasL pathway and increasing caspase 8 and 3 activation (Chen *et al.* 2002). In contrast to apoptotic mechanisms of adult *S. mansoni*, which was B cell-dependent, elimination of B cells could not reduce apoptosis in T cells exposed to schistosomula, whereas blocking Fas receptor prevented this event, indicating Fas-dependent pathway in T cells mediates apoptosis.

Given the pivotal function of T cell in amplification of anti-parasite response through the release of inflammatory mediators and exciting other immune cells, suppressing adaptive immunity via apoptosis of such critical immune cells can provide a secure environment for the early stage of infection.

NEMATODES

Trichinella spiralis

Trichinella spp. are an exceptional type of parasitic worms in terms of life cycle. In fact, this worm is able to complete all three stages of its life cycle including infective muscle larvae along with adult and newborn larvae in the same host. Trichinellosis is regarded as a food-borne disease, as consumption of the raw meat infected with larvae can evolve to infection upon release of larvae in the stomach (Gottstein *et al.* 2009). Then, the larvae penetrate into the enterocytes of the small intestine to reach the adult stage. Skeletal muscle cells are the most attractive destination for newborn larvae where they develop into muscle larvae. Immunologically, the larvae occupy a privileged environment in the muscle, as well as exploit mechanisms to transform infected muscle cells in a type of cells known as nurse cells (Sofronic-Milosavljevic *et al.* 2015). In addition, intestinal epithelial cells are another location where larvae tend to dwell for development (Gottstein *et al.* 2009).

It is well-known that some intestinal worms such as *Nippostrongylus brasiliensis* and *Trichinella spiralis* are able to trigger apoptotic mechanisms in the intestine cells, but the precise mechanism and major involved component need to be elucidated (Kuroda *et al.* 2002; Piekarska *et al.* 2009a, b). Interestingly, the worm has evolved an adaptive mechanism to up-regulate and down-regulate apoptosis-related genes in the muscle cells for providing a suitable environment in the nurse cells (Babal *et al.* 2011). *T. spiralis* is able to monitor environmental events and via the release of its ES molecules manipulates apoptosis process to sustain the longevity, accommodation and its microenvironment niche (Babal *et al.* 2011).

T. spiralis exploits mitochondrial-independent mechanisms to induce apoptosis in the murine intestine cells (Yu *et al.* 2014). During an *in vivo* study, it was revealed that in the intestine cells of the mice infected with *T. spiralis*, apoptosis is triggered via an ERS-dependent pathway. In this mechanism caspase 12 plays an essential role in triggering caspase 9 and 3 to induce cell death during infection with *T. spiralis* (Yu *et al.* 2014). In this study, it was shown that infection with *T. spiralis* causes cleavage of caspase 12 by ERS-related molecules and phosphorylation of c-Jun N-terminal protein kinase (JNK), which eventually mediates ERS-induced apoptosis (Yu *et al.* 2014). Up to now, limited data have been provided on the precise mechanism of apoptosis by *T. spiralis*. Based on the recent evidence, ERS has been found to contribute to the regulatory operation of intestinal epithelial cells and interaction between host and pathogen (McGuckin *et al.* 2010), but the main purpose of targeting ERS-related pathway by this worm to induce apoptosis remains unknown and would be of great interest in future investigations.

Filarial nematodes

Filarial nematodes invade a wide variety of animals and human. Lymphatic filariasis (LF) is one of the most prevalent diseases in tropical areas, which has provided serious problems associated with public health (Semnani and Nutman, 2004). Third stage larvae (L3) are transferred into the hosts upon the bite of the infected mosquito as a vector carrying L3. *Wuchereria bancrofti* and *Brugia malayi* are the most important causative agents responsible for LF in human (Semnani and Nutman, 2004). The female nematodes dwell in the lymphatics and release a huge amount of microfilariae distributing through blood in the peripheral circulation. Despite such a number of microfilariae in the blood vessels, the light clinical outcome emerges in patients and an immunoregulatory environment is set up by the worm whereby suppresses the potential protective Th1 and Th2 responses (Semnani and Nutman, 2004; Babu and Nutman, 2014). Such suppressive functions are attributed to the ability of the worm in forestalling DCs maturation, stimulation of alternative activation macrophages, inhibition of toll-like receptors (TLRs) expression on APCs and induction of apoptosis in various immune cells such as monocytes, DCs, NKs and T cells (Semnani and Nutman, 2004).

Brugia pahangi secretes antigens (BpA) that induce apoptosis in human monocytes, as well as forestall the proliferation of phytohemagglutinin (PHA)-treated human T cells. Although caspase 3 is involved in BpA-mediated apoptosis, the exact mechanism by which BpA trigger apoptosis in monocytes is unknown yet (Das Mohapatra *et al.* 2014).

However, it is believed that the apoptotic effect of BpA is likely mediated through TLR4.

Silencing TLR4 in monocytes was found to diminish the apoptotic effect of BpA, while overexpression of TLR4 facilitates BpA-mediated apoptosis, suggesting that TLR4 plays an important role in monocytes undergoing apoptosis. Interestingly, monocyte derived from filarial-infected humans showed resistance to apoptosis by BpA, indicating chronic infection possibly has removed susceptible monocytes and resistant ones have survived (Das Mohapatra *et al.* 2014). Live microfilariae (mf) of *B. malayi* and L3 larvae have been shown to affect the function of human NK cells through a perplexing mechanism. Both of them are able to activate NK cells to produce IFN- γ and TNF- α , as well as live microfilaria stimulate NK cells to express Th2-associated cytokines such as IL-4 and IL-5 (Babu *et al.* 2007). Interestingly, it has been found that L3 larvae after stimulation of NK cells, induce apoptosis in these cells via the caspase-dependent pathway, indicating a possible mechanism to curb host immunity. But, it is unknown why L3 larvae at first instigate NK cells to produce Th1-associated cytokines, subsequently eliminate these cells through apoptosis (Babu *et al.* 2007).

Apart from NK cells, live mf restricts anti-parasite innate responses at the first step of infection via activating the apoptotic pathway in DCs (Semnani *et al.* 2008). Exposure of human DCs and macrophages to live microfilaria of *B. malayi* showed that DCs significantly undergo apoptosis, while macrophages are resistant to microfilaria-induced apoptosis. It was revealed that mf through up-regulation of apoptosis-associated genes such as TRAIL and TNF- α stimulating cytochrome c and caspase 9, induces cell death in DCs (Semnani *et al.* 2008). However, no explanation has been provided to explain why macrophages can resist to mf-induced apoptosis and it needs further experiments to be elucidated.

It has only recently become apparent that *B. malayi* mf is able to induce another type of cell death in human DCs known as autophagy, which is a self-degradative process of cytosolic components occurred in the special occasions such as nutrient stress (Narasimhan *et al.* 2016). Molecular dissection with exploiting global proteomic analysis suggests that mammalian target of the rapamycin (mTOR) pathway as the key signalling pathway in the orchestration of autophagy is downregulated by mf. In fact, authors suppose that mf releases biomolecules targeting metabolomic pathways in DC, which eventually affect mTOR signalling to induce autophagy (Narasimhan *et al.* 2016).

Elimination of T cells as the most important cells in the orchestration of anti-parasite response is the next goal of microfilaria to exhaust adaptive immunity. CD4⁺ T cells are widely targeted by microfilaria

to be depleted through apoptosis mechanism (Jenson *et al.* 2002). Suppression and apoptosis of spleen-derived mice lymphocytes after culturing with *B. pahangi* mf have ascribed to an indirect mechanism through which mf activate the apoptotic pathway in CD4⁺ T cells via increase of IFN γ secretion and nitric oxide production (Jenson *et al.* 2002). It is hypothesized that all of these events ultimately cause CD4⁺ T cells to be susceptible to cell death. Elimination of CD4⁺ T cells by *Wuchereria bancrofti* through targeting peripheral B-1 cells has recently been shown as a potential mechanism to induce hypo-responsiveness and increase of IL-10 in infected patients (Mishra *et al.* 2017). A positive correlation between the expression of FasL (death ligand) in B-1 cells and apoptosis of Th cells in infected patients shows that B-1 cells are likely the most important orchestrator of immune anergy and immunosuppression during filariasis (Mishra *et al.* 2017).

Hartmann *et al.* (2013) during an *in vitro* study examined the apoptotic effects of two *Onchocerca volvulus*-derived recombinant proteins named abundant larval transcript-2 (OvALT-2) and novel larval transcript-1 (OvNLT-1). Exposure of ovalbumin-specific CD4⁺ DO11.10, OT-II T cells, and CD8⁺ OT-IT cells to OvALT-2 OvNLT-1 resulted in the suppression of DNA synthesis, cell division and cytokine production such as IL-2 and IFN γ from CD8⁺ OT-IT (Hartmann *et al.* 2013).

Dirofilaria immitis is a less well-known filarial nematode responsible for canine heartworm disease. It involves the cardiopulmonary system of canids after dwelling in the pulmonary arteries. The immunosuppressive function of this nematode is unknown, but it has been reported that leucocytes of dogs naturally infected with *D. immitis* significantly undergo apoptosis (Dimri *et al.* 2012). However, it has been found that the level of oxidative status has a significant correlation with apoptosis, but the main mechanism of apoptosis mediated by this worm has not been revealed (Dimri *et al.* 2012).

CESTODES

Taenia spp.

Taenia solium and *Taenia crassiceps* are the most well-known tapeworms as their immunopathogenesis has more been investigated and they are responsible for neurocysticercosis and taeniasis in humans and canine, respectively (Gonzales *et al.* 2016). Embryonated eggs of *T. solium* upon ingestion are located in the intestine, then penetrate into intestine wall and reach the systemic circulation in the body through blood vessels (Fleury *et al.* 2016). The adult tapeworms reside in the human intestine and cause intestinal taeniasis, whereas cystic larvae

(cysticercus) invade human nervous system and establish an asymptomatic infection known as neurocysticercosis (Gonzales *et al.* 2016). Various mechanisms have been reported to explain the longevity and development of this quiet invasion (Fleury *et al.* 2016). In this regard, parasitic cysts have been found to modulate host immunity via exploiting subtle tricks including, increase of Tregs and regulatory-associated cytokines (IL-10 and TGF- β), suppression of Th1 response and associated cytokines (IL-1 and IL-12) and in turn stimulation of Th2 response and IL-4 production (Peón *et al.* 2013; Arce-Sillas *et al.* 2016), interruption in activation of complement system (Sciutto *et al.* 2007) and induction of apoptosis in host immune cells (Tato *et al.* 2004; Solano *et al.* 2006).

Some studies have provided data suggesting *T. solium* mostly targets T lymphocytes to induce apoptosis. For example, it was shown that a compound from *T. solium* metacestode with cysteine protease activity is able to induce cell death in human CD4⁺ T lymphocytes. However, *in vitro* co-culture of living cysts with lymphocytes provided no results of apoptosis, suggesting metacestode is likely the major player in attenuation of the immune response through reduction of CD4⁺ T lymphocytes (Tato *et al.* 2004). Further experiments on the pigs with cysticercosis indicated that lymphocytes which during an inflammatory response recruited around the parasite are killed by metacestodes and cysteine proteases both in the brain and muscle (Solano *et al.* 2006; Sikasunge *et al.* 2008).

Another metacestode-derived compound, which has been proposed to induce apoptosis in human eosinophils is a novel annexin molecule known as annexin B1 (Yan *et al.* 2008). Yan *et al.* (2008) believe that the annexin B1 is able to attach to the surface of host eosinophils and activates apoptosis-related molecules such as caspase 3 and cytochrome c through induction of Ca²⁺ influx, implying the involvement of the mitochondrial pathway. It seems that release of annexin B1 by metacestodes of *T. solium* is an effective way to overcome anti-parasite immune response through killing eosinophils, which are the most important immune cells in protection against helminth infections.

Zepeda *et al.* (2010, 2017) have shown that *T. crassiceps* is capable of triggering apoptosis in both immune and non-immune cells. Early assessments during intraperitoneal injection of *T. crassiceps* metacestodes in mice indicated that 12 days post-infection the quantity of peritoneal inflammatory cells was significantly reduced (Zepeda *et al.* 2010). Using TUNEL assays, it was shown that high level of B and T lymphocytes including CD4⁺, CD8⁺ and CD19⁺ and eosinophils had been killed due to apoptosis by infection (Zepeda *et al.* 2010). Interestingly, they suggested that there is a

metacestode-derived compound in peritoneal fluid of mice infected with *T. crassiceps*, which can induce apoptosis in spleen CD4⁺ T lymphocytes. In addition, during an *ex vivo* evaluation splenocytes of the experimental mice showed a high level of TGFβ and Foxp3 expression in comparison with control cells (Zepeda *et al.* 2016). Similar *in vitro* results have been reported for apoptosis in CD4⁺ and CD19⁺ splenocytes of 30-day infected mice exposing to cysticercal antigens (Lopez-Briones *et al.* 2003). Given the critical role of Th1 lymphocytes in protection against early establishment of this infection (Peón *et al.* 2013), it seems that depletion of host CD4⁺ T cells through apoptosis is an intelligent strategy to set up and secure prolonged infection.

Echinococcal spp.

The echinococcal parasites causing hydatidosis involve the human and wide range of domestic and wild animals (Brunetti *et al.* 2011). They masterfully regulate their intermediate host immunity to set up cystic bodies, which are quietly distributed in many internal organs (Diaz *et al.* 2016). Dog and fox as the definitive hosts are infected by larval (metacestode) stages of echinococcal parasites, whereas in human and other animals they establish a chronic stage of the infection forming cysts (Brunetti *et al.* 2011; Diaz *et al.* 2016). This parasite is able to exploit various strategies to suppress immune cells and escape from recognition by host immunity including alteration and masking surface antigens, shaping cytokine profile, and interruption in antigen presentation and T cell activation (Zhang *et al.* 2008; Diaz *et al.* 2016).

One of the main mechanisms by which echinococcal parasites both survive in the host and overcome parasite threatening responses is the induction of apoptosis (Nono *et al.* 2012; Zhang *et al.* 2012). DCs have been found to be prone to cell death when exposed to excretory-secretory molecules released by *Echinococcus multilocularis* larvae (Nono *et al.* 2012). Murine DCs were co-incubated with the larval-derived material, then their viability was checked by trypan blue exclusion. The number of surviving cells was significantly decreased as compared with control group (Nono *et al.* 2012). Also, the same apoptotic effects, but stronger, were observed in DCs treated with *E. multilocularis* metacestode vesicles. To confirm, Annexin-V/7-AAD dual staining was applied to distinguish apoptosis from necrosis of DCs, which it confirmed high level of apoptosis in ES-treated DCs (Nono *et al.* 2012). Interestingly, Nono *et al.* (2012) showed that in contrast to BMDCs, which are susceptible to cell death by ES products of *E. multilocularis*, spleen cells in particular CD19⁺ (B cells) and CD192 (primarily T cells) are resistant to apoptosis. However, no information is available why these

spleen-derived immune cells are more durable than DCs in undergoing apoptosis in response to ES products. Of note, as indicated earlier, it has been shown that DCs are more susceptible to apoptosis than macrophages when exposed to live mf.

It seems that induction of apoptosis in host DCs by helminth materials is highly telling for parasite survival and that impairing DCs at the early stages of infection can effectively forestall inflammatory responses around the site of the parasite-mediated lesion.

There is solid evidence implying that hepatocytes in the early stages can up-regulate anti-apoptosis genes to encounter *E. multilocularis*, but in the late stages of infections the worm overcomes hepatocyte resistance and induces apoptosis (Zhang *et al.* 2012). The expression of genes regulating cell growth and apoptosis has simultaneously been measured in hepatocytes of infected mice (Zhang *et al.* 2012). Interestingly, it was found that at the early and middle stage of infection, ERK1/2 and downstream molecules such as CyclinD1, A, B1, Gadd45b and PCNA are upregulated in hepatocyte, amplifying both hepatocytes proliferation and anti-apoptotic response to combat tissue damage due to parasite itself and inflammatory cytokines such as TNFα (Zhang *et al.* 2012). In contrast, at the late stage of infection, JNK pathway was activated and the expression of genes supporting hepatocyte growth arrest/apoptosis such as p53, p21, Gadd45c and cleaved-caspase 3 was upregulated (Zhang *et al.* 2012). Caspase 3 is increased during the apoptosis, also TUNEL assay confirmed DNA breakage during *E. multilocularis* infection, suggesting induction of apoptosis is an efficacious mechanism at the late stage of infection. It seems that both parasite-derived toxic by-products and parasite-induced inflammation contribute synergistically in the promotion of apoptosis (Zhang *et al.* 2012).

To prolong their longevity within their hosts, these worms need to manipulate the lifespan of host immune cells. The main pathways of apoptosis in mammalian cells along with the potential helminth-associated intervention are illustrated (Fig. 1). In addition, the process of apoptosis induces an anti-inflammatory environment along with induction of an anergic state in APCs (Fig. 2). In fact, uptake of apoptotic cells by the engagement of scavenger receptors on APCs has been found to inhibit antigen presentation, production of pro-inflammatory mediators and priming adaptive immunity via the release of anti-inflammatory cytokines including TGFβ and IL-10 from APCs (Voll *et al.* 1997; Fadok *et al.* 1998; Albert, 2004).

POTENTIAL THERAPEUTIC APPLICATIONS

One of the new emerging oncotherapeutic investigations is exploiting helminths and their products as an

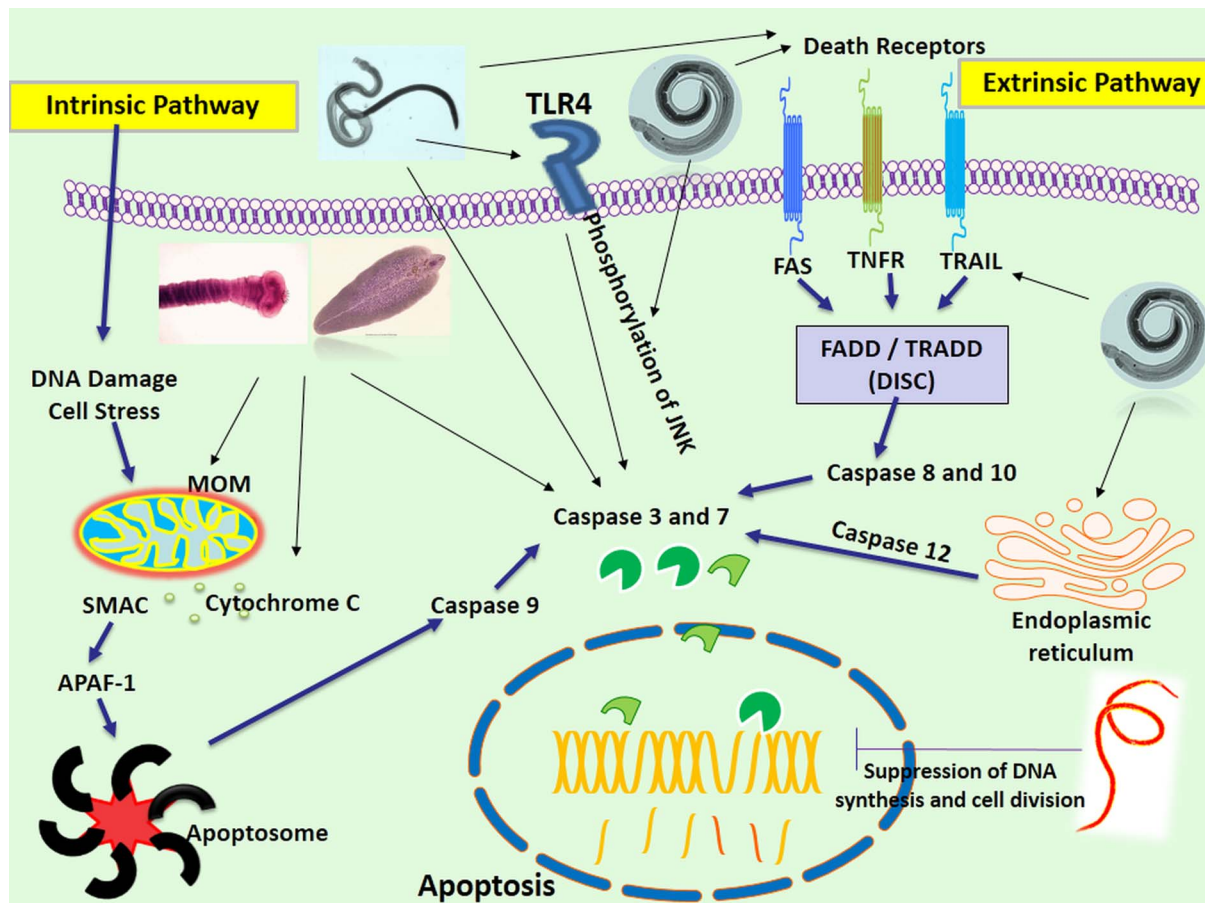


Fig. 1. An overview of the potential mechanisms of helminth-induced apoptosis. The major molecules involved in apoptosis have been illustrated and two main pathways indicated by bold arrows. The extrinsic pathway is triggered by stimulation of death receptor and mediated by the formation of DISC, which is responsible for caspases 8 and 10 activation, while the intrinsic pathway typically mediated by mitochondria upon membrane depolarization via DNA damage and cellular stress. In this pathway, mitochondria play a pivotal role in the orchestration of signalling cascade in the cytosol. Helminths can target various molecules and pathways involving in apoptosis. They are able to activate both extrinsic and intrinsic pathways directly or indirectly. For example, some of them stimulate mitochondria to release cytochrome c and SMAC leading to the formation of apoptosome and activation of caspase 3. On the other hand, some worms can elicit extrinsic pathway through stimulating death receptors and intracellular molecules resulting in DISC formation and caspase 3 activation. Interestingly, caspase 3 can also be activated through ER-mediated pathway during trichinellosis. In addition, DNA synthesis might be suppressed by *Onchocerca volvulus* as a potential mechanism to prevent and cell division. DISC, death-inducing signal complex; ER, endoplasmic reticulum; SMAC, second mitochondria-derived activator of caspases.

effective agent in priming apoptosis in cancer cells. Several *in vitro* and *in vivo* studies have conducted to display anti-cancer activity of helminth-derived biomolecules via triggering apoptosis pathway, which has been indicated in the following.

Inhibitory effects of *S. mansoni*-derived molecules on the cell proliferation show that this worm is masterful in the manipulation of cell cycle checkpoints to facilitate apoptosis process (Yang *et al.* 2013). Exposure of murine myeloid leukaemia WEHI-3B JCS cells to a recombinant protein of the worm (rSj16) provided interesting findings on the mechanisms by which rSj16 suppresses proliferation of these cells (Yang *et al.* 2013). This *in vitro* study showed that rSj16 not only inhibits the growth of the cells via suppressing G0/G1 phase, but also promotes apoptosis through targeting mitochondrial

membrane potential and an increase of caspase 3, 6, and 9 activity. Interestingly, it was revealed that rSj16 is able to up-regulate pro-apoptotic Bax expression and down-regulate anti-apoptotic Bcl-2 expression (Yang *et al.* 2013).

Recently, it has been reported that *T. spiralis* and excretory-secretory (ES L1) molecules derived from muscle larvae have inhibitory effects on the melanoma progression and the size of the tumour in mice (Vasilev *et al.* 2015). Further *in vitro* assessments showed that ES L1 antigens reduce cell proliferation and longevity of B16 melanoma cells through the increase of apoptosis. The main mechanism by which ESL1 suppresses cell proliferation is activation of the outer caspase-dependent apoptotic pathway. In support of this, treatment of melanoma cells with caspase-3 and -8 inhibitors prevented the apoptotic effects of

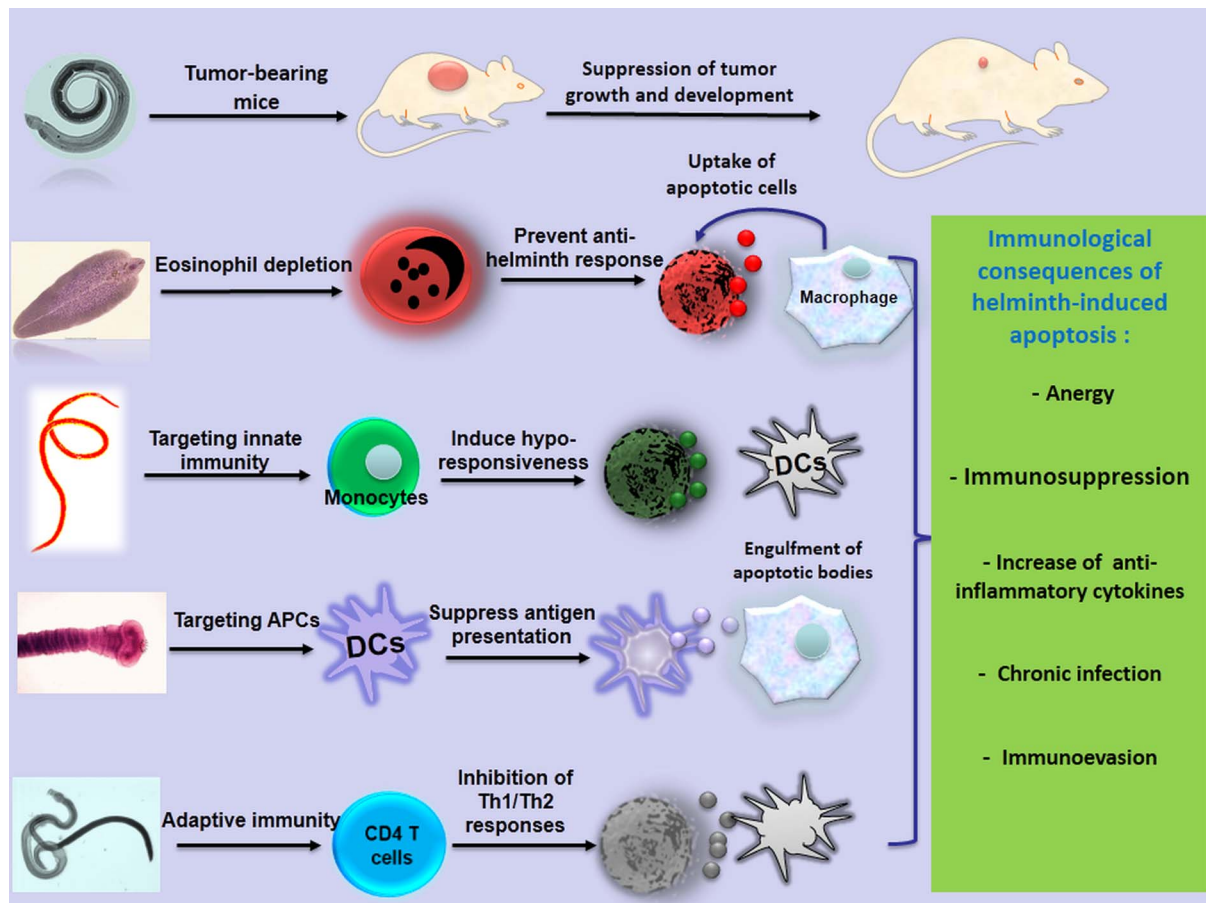


Fig. 2. A schematic illustration of direct and indirect effects of helminths and their products on host immune cells. As shown here, apoptosis occurs in various immune cells during infection with helminths. Helminth-induced apoptosis plays an essential role in parasite survival not only through suppression of anti-parasite immunity, but also via inhibition of immune-mediated tissue injury. Phagocytosis of apoptotic cells provides an immunoregulatory environment due to the release of anti-inflammatory mediators such as TGF β and IL-10 from APCs. On top of that, induction of apoptosis in certain immune cells such as CD4⁺ T cells and APCs results in hypo-responsiveness and anergy. Importantly, depletion of host immune cells paves the way to set up a chronic infection thereby guarantee transmission between various hosts. On the other hand, apoptotic functions of some helminths and their products have received a great of interest to exploit them for fighting against cancers. Although promising results have been provided in most *in vitro* studies, more *in vivo* models should be conducted to address the anti-tumour activity of helminth-derived compounds. APCs, antigen presenting cells.

ES L1, indicating death receptor is involved in ES L1-mediated apoptosis (Vasilev *et al.* 2015).

Furthermore, *T. spiralis* can forestall the development and metastasis of melanoma tumour through modulation of cytokine profile in infected mice (Kang *et al.* 2013). Results derived from cytokine array showed that infection with *T. spiralis* in tumour-bearing mice results in up-regulation of CXCL9, CXCL10 and CXCL13 in comparison with un-infected mice (Kang *et al.* 2013). However, no explanation has been provided concerning the possible chemokine-related mechanisms through which *T. spiralis* suppress tumour progress. Prospectively, the apoptosis-associated factors and their relevance with a decrease of tumour growth and metastasis need to be evaluated (Kang *et al.* 2013).

The apoptotic effects of *T. spiralis* have widely been investigated in cancerous cell lines and tumour-bearing mouse models. In this regard, Wang *et al.* have provided interesting findings on the apoptotic functions

of this worm both *in vitro* and *in vivo* (Wang *et al.* 2009, 2013). Anti-tumour activity of crude *T. spiralis* extract on five different cell lines including murine forestomach carcinoma (cell line MFC), ascetic hepatoma (cell line H22) and sarcoma (cell line S180), human chronic myeloid leukaemia (cell line K562) and hepatoma (cell line H7402) has been documented. In addition, further experiments were shown that infection of mice with viable *T. spiralis* larvae can reduce the development of murine tumours such as murine forestomach carcinoma, ascetic hepatoma and sarcoma (Wang *et al.* 2009). The apoptotic activity of *T. spiralis* antigens was also demonstrated by the construction of a recombinant protein (A200711) from the cDNA library of the worm in T7 phage display. Exposure of human hepatoma cell line (H7402) to A200711 resulted in apoptosis and inhibition of cell proliferation (Wang *et al.* 2013).

However, there are some helminth species such as *Opisthorchis viverrini*, *Clonorchis sinensis* and

Schistosoma haematobium, which are able to induce cholangiocarcinoma and urinary bladder carcinoma, in affected tissues including bile ducts and urinary bladder, respectively. Various factors can participate in the carcinogenesis of these helminths. Induction of chronic inflammation due to the release of eggs and other helminth-associated secretory products results in production of free radicals such as ROS and reactive nitrogen species, which eventually cause DNA damage (Brindley *et al.* 2015). Precisely, it has been revealed that the level of carcinogenic metabolites such as catechol estrogens and oxysterols, which are capable of affecting DNA is increased in patients suffering from opisthorchiasis and urogenital schistosomiasis (Jusakul *et al.* 2012; Gouveia *et al.* 2015). Furthermore, other factors are at play, such as N-nitroso compound formation by parasites, which is able to accelerate neoplastic transformation and DNA mutagenesis (Ohshima *et al.* 1994). Thus, the genotoxicity of these helminths is likely due to the presence of such carcinogenic metabolites and production of ROS that are augmented around the inflammation resulting in DNA damage.

One of the hallmarks of allergic asthma is severe eosinophilia in the airway lumen, which is responsible for many clinical manifestations of this disease. It has been documented that recruited eosinophils possess an unusual lifespan and reside around the bronchial wall for a long time even in the absence of allergen exposure (Felton *et al.* 2014). Thus, apoptosis appears to be an essential event affecting the resolution of eosinophil-associated airway inflammation.

It has been indicated that newly excysted metacercariae of *Paragonimus westermani* (PwNEM), a lung fluke worm, is able to release excretory-secreted products (ESP) that induce apoptosis in human eosinophils (Min *et al.* 2004). In fact, *in vitro* co-culture of human eosinophils with ESP caused phosphatidylserine (PS) externalization on the outer surface of eosinophils along with caspase3 activation, leading to parasite evasion and suppression of eosinophil-mediated local inflammation (Min *et al.* 2004).

However, it is well known that some helminth-derived products have significant apoptotic effects on eosinophils (Min *et al.* 2004; O'Connell and Nutman, 2015; Huang and Appleton, 2016), but no proof still exists concerning the helminth-mediated suppression of allergic asthma via eosinophil apoptosis. Thus, in the future studies, it would be of interest to seek whether exploiting such natural biomolecules may represent a significant step forward against asthma, as well as other eosinophil-mediated disorders.

Concluding remarks and future directions

Apart from apoptosis, several other mechanisms have been recognized thereby helminths arrest

immunity and prolong their infections including an increase of immunoregulatory cells (Maizels and McSorley, 2016), inhibition of Th1 or Th2 responses (Obieglo *et al.* 2016; Valanparambil *et al.* 2017) and targeting pattern recognition receptors, especially TLRs (Zakeri *et al.* 2016).

So far, helminth-induced activation of apoptotic pathways has only been partially studied and the molecular entities orchestrating these pathways during an acute and chronic infection remain to be elucidated. Thus, our understanding of the precise mechanisms involved in helminth-induced apoptosis and their significance clearly lags. Recognition of main worm-derived compound involved in host cell death not only can be useful for future researches making a profound understanding of worm–host interaction and anti-parasite response, but also offers novel potential therapeutics against life-threatening diseases such as cancer. However, it requires more *in vivo* and preclinical studies to examine its feasibility and applicability in a real situation. The main question that principally needs to be addressed is whether the immune system is suppressed during anti-tumour activity of helminths or not.

With exploiting omics-based technologies such as proteomics and genomics considerable progress being made to identify the main helminth-derived compounds and characterize the profile of released exosomes responsible for anergy induction and hyporesponsiveness in host immune cells via targeting apoptosis pathway.

An interesting issue that needs to be addressed is whether neutralizing the main pathways and death receptors by which helminths exert immune cell death will be helpful to elevate the potency of early immune responses and combat helminth infections. Here just the molecular process of helminth-induced apoptosis as a less-known modality of their pathogenicity has been discussed. It is also quite unknown whether other forms of cell death, except apoptosis, including necrosis, autophagy and pyroptosis can be exerted by helminths to create an immune privilege area around them. Of note, it should be taken into account that it is not always possible to distinguish activation-induced cell death driven by strong immune activation, from apoptosis actively being driven by the parasite in an attempt to directly dampen immune reactivity. Thus, in many of the papers published over the years the authors have used the word 'apoptosis' when they have really not distinguished between apoptosis, necrosis or indeed pyroptosis.

Generally, induction of apoptosis is not restricted to helminths, it contributes to the pathogenesis of many parasitic infections as an effective tool to dampen host immunity and ensure survival. For example, several protozoan-mediated infections including toxoplasmosis, leishmaniasis and malaria exploit apoptosis to conceal their infection through

the sophisticated mechanisms. Collectively, these findings provide evidence for such an orphan aspect of host–parasite interplay, which is subject to bypass host immunity by the parasitic worms. It appears to be attractive in terms of parasite longevity, immunosuppression and evasion with implications beyond parasitology.

ACKNOWLEDGEMENTS

The author is heartedly grateful to Professor Rick Maizels (Wellcome Centre for Molecular Parasitology, Institute of Infection, Immunity and Inflammation, University of Glasgow, UK) for critical and insightful comments, careful review and editing the manuscript.

FINANCIAL SUPPORT

This research received no specific grant from any funding agency, commercial or not-for-profit sectors.

REFERENCES

- Albert, M. L. (2004). Death-defying immunity: do apoptotic cells influence antigen processing and presentation? *Nature Reviews Immunology* **4**, 223–231. doi: 10.1038/nri11308.
- Arce-Sillas, A., Alvarez-Luquin, D. D., Cardenas, G., Casanova-Hernandez, D., Fragoso, G., Hernandez, M., Proano Narvaez, J. V., Garcia-Vazquez, F., Fleury, A., Sciutto, E. and Adalid-Peralta, L. (2016). Interleukin 10 and dendritic cells are the main suppression mediators of regulatory T cells in human neurocysticercosis. *Clinical & Experimental Immunology* **183**, 271–279. doi: 10.1111/cei.12709.
- Babal, P., Milcheva, R., Petkova, S., Janega, P. and Hurnikova, Z. (2011). Apoptosis as the adaptation mechanism in survival of *Trichinella spiralis* in the host. *Parasitology Research* **109**, 997–1002. doi: 10.1007/s00436-011-2343-2.
- Babu, S. and Nutman, T. B. (2012). Immunopathogenesis of lymphatic filarial disease. *Seminars in Immunopathology* **34**, 847–861. doi: 10.1007/s00281-012-0346-4.
- Babu, S. and Nutman, T. B. (2014). Immunology of lymphatic filariasis. *Parasite Immunology* **36**, 338–346. doi: 10.1111/pim.12081.
- Babu, S., Blauvelt, C. P. and Nutman, T. B. (2007). Filarial parasites induce NK cell activation, type 1 and type 2 cytokine secretion, and subsequent apoptotic cell death. *Journal of Immunology* **179**, 2445–2456.
- Bai, L. and Wang, S. (2014). Targeting apoptosis pathways for new cancer therapeutics. *Annual Review of Medicine* **65**, 139–155. doi: 10.1146/annurev-med-010713-141310.
- Barsoum, R. S., Esmat, G. and El-Baz, T. (2013). Human schistosomiasis: clinical perspective: review. *Journal of Advanced Research* **4**, 433–444. doi: 10.1016/j.jare.2013.01.005.
- Bienvenu, A. L., Gonzalez-Rey, E. and Picot, S. (2010). Apoptosis induced by parasitic diseases. *Parasites & Vectors* **3**, 106. doi: 10.1186/1756-3305-3-106.
- Brindley, P. J., da Costa, J. M. C. and Sripa, B. (2015). Why does infection with some helminths cause cancer? *Trends in Cancer* **1**, 174–182.
- Brunetti, E., Garcia, H. H., Junghanss, T. and International Cystic Echinococcosis Workshop in Lima, P. (2011). Cystic echinococcosis: chronic, complex, and still neglected. *PLOS Neglected Tropical Diseases* **5**, e1146. doi: 10.1371/journal.pntd.0001146.
- Burke, M. L., Jones, M. K., Gobert, G. N., Li, Y. S., Ellis, M. K. and McManus, D. P. (2009). Immunopathogenesis of human schistosomiasis. *Parasite Immunology* **31**, 163–176. doi: 10.1111/j.1365-3024.2009.01098.x.
- Carneiro-Santos, P., Martins-Filho, O., Alves-Oliveira, L. F., Silveira, A. M., Coura-Filho, P., Viana, I. R., Wilson, R. A. and Correa-Oliveira, R. (2000). Apoptosis: a mechanism of immunoregulation during human *Schistosomiasis mansoni*. *Parasite Immunology* **22**, 267–277.
- Carrero, J. A. and Unanue, E. R. (2006). Lymphocyte apoptosis as an immune subversion strategy of microbial pathogens. *Trends in Immunology* **27**, 497–503. doi: 10.1016/j.it.2006.09.005.
- Chen, K. M., Lee, H. H., Lai, S. C., Hsu, L. S., Wang, C. J. and Liu, J. Y. (2008). Apoptosis in meningoencephalitis of *Angiostrongylus cantonensis*-infected mice. *Experimental Parasitology* **119**, 385–390. doi: 10.1016/j.exppara.2008.03.013.
- Chen, L., Rao, K. V., He, Y. X. and Ramaswamy, K. (2002). Skin-stage schistosomula of *Schistosoma mansoni* produce an apoptosis-inducing factor that can cause apoptosis of T cells. *Journal of Biological Chemistry* **277**, 34329–34335. doi: 10.1074/jbc.M201344200.
- Chow, S. C., Brown, A. and Pritchard, D. (2000). The human hookworm pathogen *Necator americanus* induces apoptosis in T lymphocytes. *Parasite Immunology* **22**, 21–29.
- Cliffe, L. J., Potten, C. S., Booth, C. E. and Grecis, R. K. (2007). An increase in epithelial cell apoptosis is associated with chronic intestinal nematode infection. *Infection and Immunity* **75**, 1556–1564. doi: 10.1128/IAI.01375-06.
- Cwiklinski, K., O'Neill, S. M., Donnelly, S. and Dalton, J. P. (2016). A prospective view of animal and human Fasciolosis. *Parasite Immunology* **38**, 558–568. doi: 10.1111/pim.12343.
- Das Mohapatra, A., Panda, S. K., Pradhan, A. K., Prusty, B. K., Satapathy, A. K. and Ravindran, B. (2014). Filarial antigens mediate apoptosis of human monocytes through Toll-like receptor 4. *Journal of Infectious Diseases* **210**, 1133–1144. doi: 10.1093/infdis/jiu208.
- Devitt, A. and Marshall, L. J. (2011). The innate immune system and the clearance of apoptotic cells. *Journal of Leukocyte Biology* **90**, 447–457. doi: 10.1189/jlb.0211095.
- Diaz, A., Casaravilla, C., Barrios, A. A. and Ferreira, A. M. (2016). Parasite molecules and host responses in cystic echinococcosis. *Parasite Immunology* **38**, 193–205. doi: 10.1111/pim.12282.
- Dimri, U., Singh, S. K., Sharma, M. C., Behera, S. K., Kumar, D. and Tiwari, P. (2012). Oxidant/antioxidant balance, minerals status and apoptosis in peripheral blood of dogs naturally infected with *Dirofilaria immitis*. *Research in Veterinary Science* **93**, 296–299. doi: 10.1016/j.rvsc.2011.04.022.
- Duan, Y., Gu, X., Zhu, D., Sun, W., Chen, J., Feng, J., Song, K., Xu, F., He, X. and He, X. (2014). *Schistosoma japonicum* soluble egg antigens induce apoptosis and inhibit activation of hepatic stellate cells: a possible molecular mechanism. *International Journal for Parasitology* **44**, 217–224. doi: 10.1016/j.ijpara.2013.11.003.
- Escamilla, A., Bautista, M. J., Zafra, R., Pacheco, I. L., Ruiz, M. T., Martinez-Cruz, S., Mendez, A., Martinez-Moreno, A., Molina-Hernandez, V. and Perez, J. (2016). *Fasciola hepatica* induces eosinophil apoptosis in the migratory and biliary stages of infection in sheep. *Veterinary Parasitology* **216**, 84–88. doi: 10.1016/j.vetpar.2015.12.013.
- Fadok, V. A., Bratton, D. L., Konowal, A., Freed, P. W., Westcott, J. Y. and Henson, P. M. (1998). Macrophages that have ingested apoptotic cells *in vitro* inhibit proinflammatory cytokine production through autocrine/paracrine mechanisms involving TGF-beta, PGE2, and PAF. *Journal of Clinical Investigation* **101**, 890–898. doi: 10.1172/JCI11112.
- Felton, J. M., Lucas, C. D., Rossi, A. G. and Dransfield, I. (2014). Eosinophils in the lung – modulating apoptosis and efferocytosis in airway inflammation. *Frontiers in Immunology* **5**, 302. doi: 10.3389/fimmu.2014.00302.
- Fernald, K. and Kurokawa, M. (2013). Evading apoptosis in cancer. *Trends in Cell Biology* **23**, 620–633. doi: 10.1016/j.tcb.2013.07.006.
- Fleury, A., Cardenas, G., Adalid-Peralta, L., Fragoso, G. and Sciutto, E. (2016). Immunopathology in *Taenia solium* neurocysticercosis. *Parasite Immunology* **38**, 147–157. doi: 10.1111/pim.12299.
- Gazzinelli-Guimaraes, P. H., Souza-Fagundes, E. M., Cancado, G. G., Martins, V. G., Dhom-Lemos, L. C., Ricci, N. D., Fiuza, J. A., Bueno, L. L., Miranda, R. R., Guatimosim, S., Gazzinelli, A., Correa-Oliveira, R., Bartholomeu, D. C. and Fujiwara, R. T. (2013). Cell apoptosis induced by hookworm antigens: a strategy of immunomodulation. *Frontiers in Bioscience (Elite Ed)* **5**, 662–675.
- Gonzales, I., Rivera, J. T., Garcia, H. H. and Cysticercosis Working Group in P. (2016). Pathogenesis of *Taenia solium* taeniasis and cysticercosis. *Parasite Immunology* **38**, 136–146. doi: 10.1111/pim.12307.
- Gottstein, B., Pozio, E. and Nöckler, K. (2009). Epidemiology, diagnosis, treatment, and control of trichinellosis. *Clinical microbiology reviews* **22**, 127–145.
- Gouveia, M. J., Santos, J., Brindley, P. J., Rinaldi, G., Lopes, C., Santos, L. L., da Costa, J. M. and Vale, N. (2015). Estrogen-like metabolites and DNA-adducts in urogenital schistosomiasis-associated bladder cancer. *Cancer Letters* **359**, 226–232.
- Guasconi, L., Serradell, M. C. and Masih, D. T. (2012). *Fasciola hepatica* products induce apoptosis of peritoneal macrophages. *Veterinary Immunology and Immunopathology* **148**, 359–363. doi: 10.1016/j.vetimm.2012.06.022.
- Hartmann, W., Brenz, Y., Kingsley, M. T., Ajonina-Ekoti, I., Brattig, N. W., Liebau, E. and Breloer, M. (2013). Nematode-derived proteins suppress proliferation and cytokine production of antigen-

- specific T cells via induction of cell death. *PLoS ONE* **8**, e68380. doi: 10.1371/journal.pone.0068380.
- Herold, S., Ludwig, S., Pleschka, S. and Wolff, T.** (2012). Apoptosis signaling in influenza virus propagation, innate host defense, and lung injury. *Journal of Leukocyte Biology* **92**, 75–82. doi: 10.1189/jlb.1011530.
- Huang, L. and Appleton, J. A.** (2016). Eosinophils in Helminth infection: defenders and dupes. *Trends in Parasitology* **32**, 798–807. doi: 10.1016/j.pt.2016.05.004.
- Jenson, J.S., O'Connor, R., Osborne, J. and Devaney, E.** (2002). Infection with *Brugia* microfilariae induces apoptosis of CD4(+) T lymphocytes: a mechanism of immune unresponsiveness in filariasis. *European Journal of Immunology* **32**, 858–867. doi: 10.1002/1521-4141(200203)32:3<60;858::AID-IMMU858><60;858::AID-IMMU858>3.0.CO;2-E.
- Jusakul, A., Loilome, W., Namwat, N., Haigh, W.G., Kuver, R., Dechakhamphu, S., Sukontawarin, P., Pinaor, S., Lee, S.P. and Yongvanit, P.** (2012). Liver fluke-induced hepatic oxysterols stimulate DNA damage and apoptosis in cultured human cholangiocytes. *Mutation Research/Fundamental and Molecular Mechanisms of Mutagenesis* **731**, 48–57.
- Kang, Y.J., Jo, J.O., Cho, M.K., Yu, H.S., Leem, S.H., Song, K.S., Ock, M.S. and Cha, H.J.** (2013). *Trichinella spiralis* infection reduces tumor growth and metastasis of B16-F10 melanoma cells. *Veterinary Parasitology* **196**, 106–113. doi: 10.1016/j.vetpar.2013.02.021.
- Kuroda, A., Uchikawa, R., Matsuda, S., Yamada, M., Tegoshi, T. and Arizono, N.** (2002). Up-regulation of Fas (CD95) and induction of apoptosis in intestinal epithelial cells by nematode-derived molecules. *Infection and Immunity* **70**, 4002–4008.
- Kushwah, R. and Hu, J.** (2010). Dendritic cell apoptosis: regulation of tolerance versus immunity. *Journal of Immunology* **185**, 795–802. doi: 10.4049/jimmunol.1000325.
- Lopez-Briones, S., Sciutto, E., Ventura, J.L., Zentella, A. and Fragoso, G.** (2003). CD4+ and CD19+ splenocytes undergo apoptosis during an experimental murine infection with *Taenia crassiceps*. *Parasitology Research* **90**, 157–163. doi: 10.1007/s00436-003-0829-2.
- Luder, C.G., Gross, U. and Lopes, M.F.** (2001). Intracellular protozoan parasites and apoptosis: diverse strategies to modulate parasite-host interactions. *Trends in Parasitology* **17**, 480–486.
- Lundy, S.K. and Boros, D.L.** (2002). Fas ligand-expressing B-1a lymphocytes mediate CD4+-T-cell apoptosis during schistosomal infection: induction by interleukin 4 (IL-4) and IL-10. *Infection and Immunity* **70**, 812–819.
- Lundy, S.K., Lerman, S.P. and Boros, D.L.** (2001). Soluble egg antigen-stimulated T helper lymphocyte apoptosis and evidence for cell death mediated by FasL+ T and B cells during murine *Schistosoma mansoni* infection. *Infection and Immunity* **69**, 271–280.
- Maizels, R.M. and McSorley, H.J.** (2016). Regulation of the host immune system by helminth parasites. *Journal of Allergy and Clinical Immunology* **138**, 666–675. doi: 10.1016/j.jaci.2016.07.007.
- McGuckin, M.A., Eri, R.D., Das, I., Lourie, R. and Florin, T.H.** (2010). ER stress and the unfolded protein response in intestinal inflammation. *American Journal of Physiology. Gastrointestinal and Liver Physiology* **298**, G820–832. doi: 10.1152/ajpgi.00063.2010.
- Min, D.Y., Lee, Y.A., Ryu, J.S., Ahn, M.H., Chung, Y.B., Sim, S. and Shin, M.H.** (2004). Caspase-3-mediated apoptosis of human eosinophils by the tissue-invasive helminth *Paragonimus westermani*. *International Archives of Allergy and Immunology* **133**, 357–364. doi: 10.1159/000077355.
- Mishra, R., Panda, S.K., Sahoo, P.K., Bal and Satapathy, A.K.** (2017). Increased Fas ligand expression of peripheral B-1 cells correlated with CD4+ T cell apoptosis in filarial infected patients. *Parasite Immunology* **39**, e12421. doi: 10.1111/pim.12421.
- Moreau, E. and Chauvin, A.** (2010). Immunity against helminths: interactions with the host and the intercurrent infections. *Journal of Biomedicine and Biotechnology* **2010**, 428593. doi: 10.1155/2010/428593.
- Narasimhan, P.B., Bennuru, S., Meng, Z., Cotton, R.N., Elliott, K.R., Ganesan, S., McDonald-Fleming, R., Veenstra, T.D., Nutman, T.B. and Semnani, R.T.** (2016). Microfilariae of *Brugia malayi* inhibit the mTOR pathway and induce autophagy in human dendritic cells. *Infection and Immunity* **84**, 2463–2472.
- Nono, J.K., Pletinckx, K., Lutz, M.B. and Brehm, K.** (2012). Excretory/secretory-products of *Echinococcus multilocularis* larvae induce apoptosis and tolerogenic properties in dendritic cells *in vitro*. *PLOS Neglected Tropical Diseases* **6**, e1516. doi: 10.1371/journal.pntd.0001516.
- Obieglo, K., Feng, X., Bollampalli, V.P., Dellacasa-Lindberg, I., Classon, C., Osterblad, M., Helmy, H., Hewitson, J.P., Maizels, R.M., Gigliotti Rothfuchs, A. and Nylen, S.** (2016). Chronic gastrointestinal nematode infection mutes immune responses to mycobacterial infection distal to the gut. *Journal of Immunology* **196**, 2262–2271. doi: 10.4049/jimmunol.1500970.
- O'Connell, E.M. and Nutman, T.B.** (2015). Eosinophilia in infectious diseases. *Immunology and Allergy Clinics of North America* **35**, 493–522. doi: 10.1016/j.iac.2015.05.003.
- O'Connor, R.A., Jenson, J.S., Osborne, J. and Devaney, E.** (2003). An enduring association? Microfilariae and immunosuppression [correction of immunosuppression] in lymphatic filariasis. *Trends in Parasitology* **19**, 565–570.
- Ohshima, H., Bandaletova, T.Y., Brouet, I., Bartsch, H., Kirby, G., Ogunbiyi, F., Vatanasapt, V. and Pipitgool, V.** (1994). Increased nitrosamine and nitrate biosynthesis mediated by nitric oxide synthase induced in hamsters infected with liver fluke (*Opisthorchis viverrini*). *Carcinogenesis* **15**, 271–275.
- Opferman, J.T.** (2008). Apoptosis in the development of the immune system. *Cell Death & Differentiation* **15**, 234–242. doi: 10.1038/sj.cdd.4402182.
- Pearce, E.J. and MacDonald, A.S.** (2002). The immunobiology of schistosomiasis. *Nature Reviews Immunology* **2**, 499–511. doi: 10.1038/nri843.
- Peón, A.N., Espinoza-Jiménez, A. and Terrazas, L.I.** (2013). Immunoregulation by *Taenia crassiceps* and its antigens. *BioMed Research International* **2013**, 498583. doi: 10.1155/2013/498583.
- Piekarska, J., Michalski, A., Szczyпка, M. and Obminska-Mrukowicz, B.** (2009a). *Trichinella spiralis*: effect of thymus factor X on apoptosis and necrosis in mice. *Experimental Parasitology* **123**, 128–133. doi: 10.1016/j.exppara.2009.06.009.
- Piekarska, J., Szczyпка, M., Obminska-Mrukowicz, B. and Gorczykowski, M.** (2009b). Effect of phytohaemagglutinin-P on apoptosis and necrosis in *Trichinella spiralis* infected mice. *Veterinary Parasitology* **159**, 240–244. doi: 10.1016/j.vetpar.2008.10.050.
- Prendergast, C.T., Sanin, D.E., Cook, P.C. and Mountford, A.P.** (2015). CD4+ T cell hyporesponsiveness after repeated exposure to *Schistosoma mansoni* larvae is dependent upon interleukin-10. *Infection and Immunity* **83**, 1418–1430. doi: 10.1128/IAI.02831-14.
- Robinson, M.W. and Dalton, J.P.** (2009). Zoonotic helminth infections with particular emphasis on fasciolosis and other trematodiasis. *Philosophical transactions of the Royal Society of London. Series B, Biological Sciences* **364**, 2763–2776. doi: 10.1098/rstb.2009.0089.
- Sciutto, E., Chavarria, A., Fragoso, G., Fleury, A. and Larralde, C.** (2007). The immune response in *Taenia solium* cysticercosis: protection and injury. *Parasite Immunology* **29**, 621–636. doi: 10.1111/j.1365-3024.2007.00967.x.
- Semnani, R.T. and Nutman, T.B.** (2004). Toward an understanding of the interaction between filarial parasites and host antigen-presenting cells. *Immunological Reviews* **201**, 127–138. doi: 10.1111/j.0105-2896.2004.00196.x.
- Semnani, R.T., Venugopal, P.G., Mahapatra, L., Skinner, J.A., Meylan, F., Chien, D., Dorward, D.W., Chaussabel, D., Siegel, R.M. and Nutman, T.B.** (2008). Induction of TRAIL- and TNF-alpha-dependent apoptosis in human monocyte-derived dendritic cells by microfilariae of *Brugia malayi*. *Journal of Immunology* **181**, 7081–7089.
- Serradell, M.C., Guasconi, L., Cervi, L., Chiapello, L.S. and Masih, D.T.** (2007). Excretory-secretory products from *Fasciola hepatica* induce eosinophil apoptosis by a caspase-dependent mechanism. *Veterinary Immunology and Immunopathology* **117**, 197–208. doi: 10.1016/j.vetimm.2007.03.007.
- Serradell, M.C., Guasconi, L. and Masih, D.T.** (2009). Involvement of a mitochondrial pathway and key role of hydrogen peroxide during eosinophil apoptosis induced by excretory-secretory products from *Fasciola hepatica*. *Molecular and Biochemical Parasitology* **163**, 95–106. doi: 10.1016/j.molbiopara.2008.10.005.
- Sikasunge, C.S., Phiri, I.K., Johansen, M.V., Willingham, A.L., 3rd and Leifsson, P.S.** (2008). Host-cell apoptosis in *Taenia solium*-induced brain granulomas in naturally infected pigs. *Parasitology* **135**, 1237–1242. doi: 10.1017/S0031182008004678.
- Smith, P., Walsh, C.M., Mangan, N.E., Fallon, R.E., Sayers, J.R., McKenzie, A.N. and Fallon, P.G.** (2004). *Schistosoma mansoni* worms induce anergy of T cells via selective up-regulation of programmed death ligand 1 on macrophages. *Journal of Immunology* **173**, 1240–1248.
- Sofronic-Milosavljevic, L., Ilic, N., Pinelli, E. and Gruden-Movsesijan, A.** (2015). Secretory products of *Trichinella spiralis* muscle larvae and immunomodulation: implication for autoimmune diseases, allergies, and malignancies. *Journal of Immunology Research* **2015**, 523875. doi: 10.1155/2015/523875.
- Solano, S., Cortes, I.M., Copitin, N.I., Tato, P. and Molinari, J.L.** (2006). Lymphocyte apoptosis in the inflammatory reaction around *Taenia solium* metacystodes in porcine cysticercosis. *Veterinary Parasitology* **140**, 171–176. doi: 10.1016/j.vetpar.2006.03.006.
- Tato, P., Fernandez, A.M., Solano, S., Borgonio, V., Garrido, E., Sepulveda, J. and Molinari, J.L.** (2004). A cysteine protease from *Taenia solium* metacystodes induce apoptosis in human CD4+ T-cells. *Parasitology Research* **92**, 197–204. doi: 10.1007/s00436-003-1008-1.

- Valanparambil, R. M., Tam, M., Jardim, A., Geary, T. G. and Stevenson, M. M.** (2017). Primary *Heligmosomoides polygyrus bakeri* infection induces myeloid-derived suppressor cells that suppress CD4+ Th2 responses and promote chronic infection. *Mucosal Immunology*, **10**, 238–249. doi: 10.1038/mi.2016.36.
- Vasilev, S., Ilic, N., Gruden-Movsesijan, A., Vasilijic, S., Bosic, M. and Sofronic-Milosavljevic, L.** (2015). Necrosis and apoptosis in *Trichinella spiralis*-mediated tumour reduction. *Central European Journal of Immunology* **40**, 42–53. doi: 10.5114/ceji.2015.50832.
- Voll, R. E., Herrmann, M., Roth, E. A., Stach, C., Kalden, J. R. and Girkontaite, I.** (1997). Immunosuppressive effects of apoptotic cells. *Nature* **390**, 350–351. doi: 10.1038/37022.
- Wang, J., Xu, F., Zhu, D., Duan, Y., Chen, J., Sun, X., He, X., Li, P., Sun, W. and Feng, J.** (2014). *Schistosoma japonicum* soluble egg antigens facilitate hepatic stellate cell apoptosis by downregulating Akt expression and upregulating p53 and DR5 expression. *PLOS Neglected Tropical Diseases* **8**, e3106.
- Wang, X. L., Fu, B. Q., Yang, S. J., Wu, X. P., Cui, G. Z., Liu, M. F., Zhao, Y., Yu, Y. L., Liu, X. Y., Deng, H. K., Chen, Q. J. and Liu, M. Y.** (2009). *Trichinella spiralis*—a potential anti-tumor agent. *Veterinary Parasitology* **159**, 249–252. doi: 10.1016/j.vetpar.2008.10.052.
- Wang, X. L., Liu, M. Y., Sun, S. M., Liu, X. L., Yu, L., Wang, X. R., Chu, L. X., Rosenthal, B., Shi, H. N., Boireau, P., Wang, F., Zhao, Y. and Wu, X. P.** (2013). An anti-tumor protein produced by *Trichinella spiralis* induces apoptosis in human hepatoma H7402 cells. *Veterinary Parasitology* **194**, 186–188. doi: 10.1016/j.vetpar.2013.01.052.
- Yan, H. L., Xue, G., Mei, Q., Ding, F. X., Wang, Y. Z. and Sun, S. H.** (2008). Calcium-dependent proapoptotic effect of *Taenia solium* metacystodes annexin B1 on human eosinophils: a novel strategy to prevent host immune response. *International Journal of Biochemistry & Cell Biology* **40**, 2151–2163. doi: 10.1016/j.biocel.2008.02.018.
- Yang, F., Sun, X., Shen, J., Yu, L. P., Liang, J. Y., Zheng, H. Q. and Wu, Z. D.** (2013). A recombinant protein (rSj16) derived from *Schistosoma japonicum* induces cell cycle arrest and apoptosis of murine myeloid leukemia cells. *Parasitology Research* **112**, 1261–1272. doi: 10.1007/s00436-012-3260-8.
- Yu, Y. R., Deng, M. J., Lu, W. W., Zhang, J. S., Jia, M. Z., Huang, J. and Qi, Y. F.** (2014). Endoplasmic reticulum stress-mediated apoptosis is activated in intestines of mice with *Trichinella spiralis* infection. *Experimental Parasitology* **145**, 1–6. doi: 10.1016/j.exppara.2014.06.017.
- Zakeri, A., Borji, H. and Haghparast, A.** (2016). Interaction between helminths and toll-like receptors: possibilities and potentials for asthma therapy. *International Reviews of Immunology* **35**, 219–248. doi: 10.3109/08830185.2015.1096936.
- Zepeda, N., Solano, S., Copitin, N., Fernandez, A. M., Hernandez, L., Tato, P. and Molinari, J. L.** (2010). Decrease of peritoneal inflammatory CD4(+), CD8(+), CD19(+) lymphocytes and apoptosis of eosinophils in a murine *Taenia crassiceps* infection. *Parasitology Research* **107**, 1129–1135. doi: 10.1007/s00436-010-1980-1.
- Zepeda, N., Tirado, R., Copitin, N., Solano, S., Fernandez, A. M., Tato, P. and Molinari, J. L.** (2016). A *Taenia crassiceps* factor induces apoptosis of spleen CD4+T cells and TFG-β and Foxp3 gene expression in mice. *Journal of Helminthology* **90**, 223–231. doi: 10.1017/S0022149X1500022X.
- Zepeda, N., Solano, S., Copitin, N., Chavez, J. L., Fernandez, A. M., Garcia, F., Tato, P. and Molinari, J. L.** (2017). Apoptosis of mouse hippocampal cells induced by *Taenia crassiceps* metacystode factor. *Journal of Helminthology* **91**, 215–221. doi: 10.1017/S0022149X16000146.
- Zhang, C., Wang, J., Lu, G., Li, J., Lu, X., Mantion, G., Vuitton, D. A., Wen, H. and Lin, R.** (2012). Hepatocyte proliferation/growth arrest balance in the liver of mice during *E. multilocularis* infection: a coordinated 3-stage course. *PLoS ONE* **7**, e30127. doi: 10.1371/journal.pone.0030127.
- Zhang, W., Ross, A. G. and McManus, D. P.** (2008). Mechanisms of immunity in hydatid disease: implications for vaccine development. *Journal of Immunology* **181**, 6679–6685.