Thymosin alpha 1

INDICATION and USAGE SUMMARY

- Thymosin alpha-1 is a synthetic thymic peptide
- Modulates innate immunity (pleiotropic)\(^1,2,3\)
  - Improves Th1 immune responses and helps balance Th1/Th2
  - Promotes T cell (Tregs) differentiation and maturation
  - Decreases T-cell apoptosis
  - Improves CD3+, CD4+ and CD8+
  - Improves production of IL-1 beta, IFN-γ, IL-2, IL-3, IL-6, IL-10
  - Improves NK cell activity and TNF-alpha
  - Improves macrophages and B cells
  - Up regulates MHC Class I expression in antigen expressing cell
  - Tumor specific antigens; anti-tumor properties
  - Inhibits viral replication
  - Activates indoleamine 2,3-dioxygenase enzyme - dampens immunity
  - Improves dendritic cell tryptophan catabolism
- Antioxidant properties – improves intracellular glutathione

- **Used for clinical conditions where immune support is necessary**
  - Conditions requiring immune response modulation
  - Hepatitis B & C
  - HIV/AIDS
  - Cancer - non-small cell lung (NSCLC), hepatocellular, malignant melanoma
  - Chemotherapy adjunct
  - Chronic inflammatory conditions; autoimmunity
  - Cystic fibrosis
  - Lyme disease
  - Blocks steroid-induced apoptosis of thymocytes
  - Depressed response to vaccinations; adjunct to flu vaccine
  - Geriatric immune support
  - DiGeorge’s syndrome
- **May reduce hematological toxicity of cytotoxic drug therapies**
  - Cyclophosphamide
  - 5-fluorouracil (5FU)
  - Dacarbazine
  - Ifosfamide
- **Zadaxin™ – proprietary thymosin alpha 1 (thymalfasin) approved in 30 countries for hepatitis B and C and cancer.**
  - Phase II clinical trials US – Hepatitis B
  - Phase III clinical trials US – Hepatitis C
  - Indicated as a monotherapy or combination therapy with interferon for the treatment of chronic hepatitis B, hepatitis C and cancer.
  - Also indicated for treatment of non-small cell lung cancer (NSCLC), hepatocellular carcinoma, AIDS and malignant melanoma.
  - General dosage
    - 1.6 mg, injected SubQ, 2 times weekly for 6-12 months
    - Patients weighing < 40 kg, dosage adjusted to 40 mcg/kg, 2 times weekly.
  - May be used together with conventional antiretroviral regimens
  - Individual dosage requirements may vary based on clinical presentation

**Name(s):** Thymosin alpha-1, thymalfasin, Zadaxin™

**Sequence:**
Ac-Ser-Asp-Ala-Ala-Val-Asp-Thr-Ser-Ser-Glu-Ile-Thr-Lys-Asp-Leu-Lys-Glu-Lys-Lys-Val-Val-Glu-Glu-Ala-Glu-Asn-OH

**Molecular formula:** C129H215N33O55

**Molar Weight:** 3108.28 g/mol

**Dosage Route:** SubQ injection

---

Rapidly absorbed, peak serum within 2 hours, 2 hour half-life

Dosage:
- SubQ General Dosage
  - 1.5 mg SubQ every 3rd day
  - Treatment from 2 weeks for viral infection and 3 months or longer for HIV/cancer /Hepatitis B, C or complicated immune suppression or over-activation
  - Multiple over-lap of usage

Zadaxin™ dosage
- 1.6 mg twice weekly for 6-12 months

Overview
Thymosin alpha 1 is peptide containing 28 amino acid residues that are N-terminally acetylated and proteolytically processed from prothymosin alpha. Thymosin alpha 1 was isolated by Goldstein and coworkers from thymosin fraction 5, a mixture of peptides from calf thymus in the 1970s. It is used to improve immune responses in times of need.

Ta1 is pleiotropic – improves innate immunity when needed, down regulates immunity when not needed. Ta1 is a thymic peptide that demonstrates a profound ability to restore immune system homeostasis in different physiological and pathological conditions (i.e., viral infections, cancer, immunodeficiency, vaccination support and immumosenescence) acting as multitasking protein depending on the host state of inflammation or immune dysfunction.

Thymosin alpha 1 helps the body induce effective host-derived immune effectors and balance the Th1 / Th2 arms of immunity. These effector cells improve various immunomodulatory properties that lead to augmentation of T lymphocyte function, including modulation of interleukin-2 (IL-2), stimulation of interferon-g (IFN-g) production, induction of T lymphocyte and natural killer (NK) cells and stimulation of thymopoiesis. Ta1 has also been reported to up-regulate MHC Class I expression in antigen-presenting cells. Additionally, Ta1 down-regulates the activity of terminal deoxynucleotide transferase (TdT) in TdT1 thymocytes, suggesting a role

---

for Ta1 in thymocyte maturation. Ta1 has also been found to antagonize both
activation induced (anti-CD3) and glucocorticoid-induced thymocyte apoptosis.\textsuperscript{16} It
has also been reported that Ta1 stimulates activity of Indoleamine-2,3-Dioxygenase
(IDO), leading to an increase in FoxP3 IL-10 producing regulatory T cells. This
increase leads to feedback inhibition of cytokine production, hence dampening
immune response to prevent a pro-inflammatory cytokine storm and possibly
autoimmune phenomena.

Immune senescence, considered an aging process, has been related to a gradual
decline in thymus function and thymic hormone production.\textsuperscript{17} The lack of thymic
hormones may contribute to the decline in immune function, particularly the T cell
component. In the elderly, antibody response after vaccination is compromised
when compared to response in young. A similar diminished antibody response has
been reported in patients with end-stage renal disease (ESRD) and in hemodialysis
patients. In hemodialysis patients, this has been attributed to incompetence in T
cell-mediated immune responses.

Since thymosin alpha-1 can enhance T-cell-dependent specific antibody production,
Ta1 can help augment specific vaccine responses both in the elderly or in younger
subjects in situations in which there are suboptimal quantities of immunizing
antigen available.\textsuperscript{18}

Thymosin alpha 1 has been used to support immunity in over 3,000 patients and in
over 70 clinical studies, either as monotherapy or in conjunction with current
allopathic medicines. The lack of significant side effects with thymosin alpha 1 is in
sharp contrast to other major immune response modulators such as IFN and IL-2,
which can lead to flu-like symptoms including malaise, fever, headache, chills and
pulmonary edema (with IL-2).\textsuperscript{19}

\textbf{Zadaxin™}

Zadaxin (thymalfasin, SciClone Pharmaceuticals, China) is a thymosin alpha 1
peptide that has been evaluated for its immunomodulatory activities and related
therapeutic potential in several diseases, including chronic hepatitis B and C,
aquired immunodeficiency syndrome (AIDS), primary immunodeficiency diseases,
depressed response to vaccination, and cancer. Zadaxin is currently in Phase III
trials for the treatment of hepatitis C and in Phase II trials for hepatitis B in the US.

\textbf{Hepatitis B - Zadaxin}\textsuperscript{21}

\textsuperscript{16} Baumann CA, Badamchian M, Goldstein AL. Thymosin alpha 1 is a time and dose-dependent antagonist of
\textsuperscript{18} Ershler WB, Gravenstein S, Geloo ZS. Thymosin alpha 1 as an adjunct to influenza vaccination in the elderly. Ann NY Acad Sci.
2007;1112:375-84.
\textsuperscript{19} Lopez-Alcorocho J, Vartolome J, Cotonat T, Carreno V. Efficacy of prolonged interferonalpha treatment in chronic hepatitis B
\textsuperscript{20} Zadaxin Drug Monograph. SciClone Pharmaceuticals. www.Sciclone.com
\textsuperscript{21} Zadaxin Drug Monograph. SciClone Pharmaceuticals. www.Sciclone.com
ZADAXIN thymosin alpha 1 (thymalfasin) is indicated as a monotherapy or combination therapy with interferon for the treatment of chronic hepatitis B. Pooled analysis of 3 randomized controlled trials comprising 223 patients was performed. Thymosin alpha 1 was administered twice weekly for 6 months. Follow-up assessments were performed at 12 months after completion of treatment (see table 1 below). In multiple studies, ZADAXIN was reported to have a delayed therapeutic response 12 months or longer after completion of therapy.

<table>
<thead>
<tr>
<th>Study Reference</th>
<th>Number of Patients Treatment Groups</th>
<th>Response Rate at 12-months followup*</th>
</tr>
</thead>
<tbody>
<tr>
<td>US Phase 2</td>
<td>12 Thymosin alpha 1 (1.6 mg SQ Biw 6 mos.) 8 Placebo</td>
<td>(88%) Thymosin alpha 1 (22%) Placebo</td>
</tr>
<tr>
<td>US Phase 3</td>
<td>50 Thymosin alpha 1 (1.6 mg SQ Biw 6 mos.) 42 Placebo</td>
<td>(24%) Thymosin alpha 1 (12%) Placebo</td>
</tr>
<tr>
<td>Taiwanese Phase 3</td>
<td>51 Thymosin alpha 1 (1.6 mg SQ Biw 6 mos.) 53 no treatment</td>
<td>(37%) Thymosin alpha 1 (25%) no treatment</td>
</tr>
<tr>
<td>Pooled Data</td>
<td>118 Thymosin alpha 1 (1.6 mg SQ Biw 6 mos.) 110 Placebo or no treatment</td>
<td>(54%) Thymosin alpha 1 (99%) Placebo or no treatment</td>
</tr>
</tbody>
</table>

*Response rate is defined as the percentage of subjects who were HIV DNA and HBeAg negative at 12-months follow up.

**Hepatitis C - Zadaxin**

ZADAXIN thymosin alpha 1 (thymalfasin) is indicated as a combination therapy with interferon for the treatment of chronic hepatitis C. Pooled analysis of 2 randomized controlled trials and 1 historical controlled trial comprising 121 ZADAXIN plus interferon, or interferon treated patients, was performed. Thymosin alpha 1 was administered at least twice weekly for 6 to 12 months and interferon was administered up to three times weekly for 6 to 12 months. Follow-up assessments were performed upon completion of treatment and at 6 months after completion of treatment (see table 2 below).

---

Table 2: Efficacy of Thymosin alpha 1 therapy for Hepatitis C

<table>
<thead>
<tr>
<th>Study Reference</th>
<th>Number of Patients Treatment Groups*</th>
<th>Response Rate at End of Treatment**</th>
<th>Sustained Response Rate***</th>
</tr>
</thead>
<tbody>
<tr>
<td>US Phase 3 [6,9]</td>
<td>35 Thymosin alpha 1 + Interferon (70 ± 16 mg SQ BID 6 mos. + IFN 3 MU TID 6 mos.) 37 Interferon (IFN 3 MU TID 6 mos.) 37 Placebo</td>
<td>ALT Response (57.7%) Thymosin alpha 1 + Interferon (16.2%) Interferon (2.7%) Placebo</td>
<td>ALT Response (19.2%) Thymosin alpha 1 + Interferon (9.4%) Interferon</td>
</tr>
</tbody>
</table>

Italy Phase 2 [6,9] 15 Thymosin alpha 1 (110 mg SQ qd for 4 days then BID for 51 wks.) + IFN 3 MU on day 4 then TID for 51 wks. | Virologic Response (71.3%) Thymosin alpha 1 + Interferon | Virologic Response (40.0%) Thymosin alpha 1 + Interferon |

Italy Phase 2 [6,9] 17 Thymosin alpha 1 (10 mg SQ BID for 6 mos. + IFN 3 MU TID 6 mos.) 17 Interferon | AL** Response (70.6%) Thymosin alpha 1 + Interferon (35.3%) Interferon | AL** Response (29.4%) Thymosin alpha 1 + Interferon (17.6%) Interferon |

Posed Data [9] 67 Thymosin alpha 1 (10 mg SQ BID 6 to 12 mos. IFN 3 MU TID 6 to 12 mos.) 54 Interferon | ALT Response (44.7%) Thymosin alpha 1 + Interferon (22.2%) Interferon* | ALT Response (22.4%) Thymosin alpha 1 + Interferon (9.3%) Interferon** |

*Intention-to-treat analysis
**ALT Response Rate is defined as the percentage of subjects who had normal ALT at end of treatment. Virologic Response Rate is defined as the percentage of subjects who were HCV RNA negative at end of treatment.
***ALT Response Rate is defined as the percentage of subjects who had normal ALT at end of 6-month follow up. Virologic Response Rate is defined as the percentage of subjects who were HCV RNA negative at end of 6 months follow up. US Phase 3 sustained response includes patients treated for 6 months and responders retreated for a total of 12 months.

Cancer - Zadaxin

ZADAXIN thymosin alpha 1 (thymalfasin) is indicated as a adjuvant therapy for chemotherapy-induced immune depression, immune insufficiency and immune suppression in patients with non-small cell lung carcinoma (NSCLC), malignant melanoma, hepatocellular carcinoma (HCC), breast cancer, non-Hodgkin's lymphoma (CHOP program), colorectal cancer, head and neck cancer, leukemia's, pancreatic carcinoma, and renal cell carcinoma. Clinical studies in over 1,000 patients with various types of cancer demonstrated that thymosin alpha 1 improved immunological parameters increased tumor response rates, and improved survival and quality of life (see table 3 for selected studies). Thymosin alpha 1 was either administered for 6 months or given between chemotherapy cycles for the duration of treatment.

Table 3: Efficacy of Thymosin alpha 1 therapy for Cancer

<table>
<thead>
<tr>
<th>Study Reference</th>
<th>Number of Patients Treatment Groups</th>
<th>Clinical Outcome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Italy pilot study (HCC) [10]</td>
<td>Thymosin alpha 1 (1.6 mg SQ 1xw 6 mos) + TACE 12 TACE only</td>
<td>Statistically significant survival benefit and improvement in immunological parameters in thymosin alpha 1 treated group compared with historical controls</td>
</tr>
<tr>
<td>US Phase 3 [NSCLC primarily Stage III] [11]</td>
<td>Thymosin alpha 1 (0.9 mg/m² SQ Biw up to 12 mos) + placebo + Thymosin alpha 1 treatment followed radiation therapy</td>
<td>Recurrence-free survival (p = 0.04) greater effect in nonbulky vs. bulky tumors, p = 0.01 Median survival 9.2 vs. 32 wks Overall survival: p = 0.002</td>
</tr>
<tr>
<td>Italy Phase 2 [NSCLC, Stage II &amp; IV] [12]</td>
<td>Thymosin alpha 1 (1 mg SQ on days 8 to 11 and 15 to 18 + Ifosfamide + IFN-α 3 MIU on days 11 and 18 + 10 Ifosfamide)</td>
<td>Objective response: 65% vs. 10% Median time to progression: 18 wks vs. 7 wks (p = 0.0059) Median survival duration: 24 wks vs. 16 wks &gt;1 yr survival: 3 (33%) vs. 2 (20%) Lymphocyte count maintained vs. decreased Hematologic toxicity reduced with no grade 3/4 toxicity compared to 50% in chemotherapy group</td>
</tr>
<tr>
<td>Italy Phase 2 [Malignant Melanoma] [13]</td>
<td>Thymosin alpha 1 (1 mg SQ on days 8 to 15 + DTIC + IFN-α Cycle repeated every 4 wks for 6 times (6 mos) or until disease progression)</td>
<td>Overall response rate: 45% mean response duration: 13.5 mos</td>
</tr>
<tr>
<td>Italy Phase 2 [Malignant Melanoma] [14]</td>
<td>Thymosin alpha 2 mg SQ days 4-7 + DTIC + IL-2 Cycle repeated every 3 wks up to 6 times (approx. 4 mos) Follow-up up to 29 mos</td>
<td>Overall response rate: 36% Median time to progression: 5.5 mos Median survival: 11 mos (46% survived greater than 1 yr)</td>
</tr>
</tbody>
</table>
HIV/AIDS – Zadaxin

Both preclinical and clinical studies have shown a high degree of immune restoration from the combined administration of Zadaxin and IFN α. Thus, Zadaxin in combination with AZT and IFN α is reported to improve outcomes for immune suppressed HIV-infected patients.

Potential Side Effects and/or Contraindications

- Thymosin alpha 1 peptide given subcutaneously is reported safe and efficacious in recommended dosages.
- Since 1979, thymosin alpha-1 is well tolerated. Tα1 has demonstrated a very favorable toxicity profile in more than 3,000 individuals treated to date, including patients with hepatocellular carcinoma, non-small-cell lung cancer, melanoma, and hepatitis B and C. 27 28 29 30
- Thymosin alpha 1 has been reported to be well tolerated even in patients with decompensated liver disease, renal disease requiring hemodialysis and primary immunodeficient individuals.
- As with all injections, redness and pain at the site of injection may be present.
- Rare adverse reactions include erythema, transient muscle atrophy, polyarthralgia combined with hand edema, and rash.
- A transient increase in ALT to more than twice baseline value can occur during thymosin alpha 1 therapy. When ALT flare occurs, thymosin alpha 1 should generally be continued unless signs and symptoms of liver failure are observed.
- Use caution if administering to pregnant or nursing women.
- Do not use in individuals being deliberately immunosuppressed.

DISCLAIMER: Statements made are for educational purposes and have not been evaluated by the US Food and Drug Administration (FDA). They are not intended to diagnose, treat, cure, or prevent any disease. Peptides should only be administered by licensed and qualified health care professionals.

30 Zadaxin prescribing information SciClone Pharmaceuticals. www.scicloneinternational.com

© Copyright 2018, Scientific Performance Research, LLC’ Cincinnati, OH