

Constitutive protein content of procaspases in murine tissue

Research Article

Amie J. Dirks-Naylor*, Samir A. Kouzi

Wingate University School of Pharmacy,
28174-8301 Wingate, USA

Received 18 June 2012; Accepted 07 September 2012

Abstract: Caspases are proteases most notably involved in apoptosis and inflammation. Although mRNA content is better described, the constitutive protein content of procaspases between tissue types is not well documented. Since mRNA and protein content do not necessarily correlate, we aimed to discern protein content differences between various tissues. Protein content of procaspase-1, -8, -9, and -12 was assessed in gastrocnemius, heart, liver, and kidney. Since highly expressed in skeletal muscle, content of procaspase-12 was also analyzed in muscles with different fiber type compositions to discern any fiber type differences. Furthermore, Western analysis for procaspase-12 revealed prominent bands of ~40 kDa and ~30 kDa under basal conditions, in addition to the 50 kDa band corresponding to the full-length procaspase. Therefore, the content of these caspase-12 related species in the tissue and muscle types is also described. Results show protein content of procaspase-1, -8, -9, and -12 and caspase-12 related species differs between tissue types and do not necessarily correlate with mRNA content reported in the published literature. Procaspase-12 content in skeletal muscle may be fiber-type dependent with higher expression in more oxidative fibers. Furthermore, the 40 kDa species of caspase-12 was the dominant form of the protein in most tissues analyzed.

Keywords: Caspase • Caspase-12 • Apoptosis • Soleus • Extensor digitorum longus (EDL) • Diaphragm

© Versita Sp. z o.o.

1. Introduction

Caspases are highly conserved cysteine-dependent, aspartate-specific acid proteases most notably involved in apoptosis and inflammation, although roles in other cellular processes have been identified [1,2]. There are 15 known mammalian caspases which function in specific apoptotic and/or inflammatory pathways activated by different stimuli. Caspases reside in the cell as inactive proenzymes. Upon stimulation, the procaspase is cleaved to remove the N-terminal pro-domain and again to produce a large and small subunit which are components of the stable heterotetrameric active caspase [1,3].

While mRNA content of various procaspases between tissue types is better described, the protein content is not well documented. It has been acknowledged that the tissue distribution of mRNA content does not always correlate with protein content [4]. Thus, the aim of the study was to discern constitutive protein content differences between various murine tissues.

Protein content of apical procaspase-1,-8,-9, and -12 were assessed in murine skeletal muscle (gastrocnemius), heart, liver, and kidney. In addition to procaspase-12, two other related species are described which may be cleavage products of caspase-12 or alternative isoforms. Moreover, since procaspase-12 is highly expressed in skeletal muscle, we aimed to determine muscle fiber type differences. Thus, we assessed the protein content of procaspase-12 and related species in soleus, extensor digitorum longus (EDL), and diaphragm; skeletal muscles with varying fiber type composition.

2. Experimental Procedures

Twelve-week old male BALB/c mice were purchased from Charles River (Wilmington, Ma). All procedures were approved by the institutional Research Review Board (number of approval: RRB-112010NAYLOR). Mice were sacrificed *via* cervical dislocation after

* E-mail: anaylor@wingate.edu