Contrasting Roles of Different Endoglin Forms in Atherosclerosis

Young-Saeng Jang and In-Hong Choi*
Department of Microbiology, Institute for Immunology and Immunological Diseases, Yonsei University College of Medicine, Seoul 120-749, Korea

Endoglin (also known as CD105 or TGF-β type III receptor) is a co-receptor involved in TGF-β signaling. In atherosclerosis, TGF-β signaling is crucial in regulating disease progression owing to its anti-inflammatory effects as well as its inhibitory effects on smooth muscle cell proliferation and migration. Endoglin is a regulator of TGF-β signaling, but its role in atherosclerosis has yet to be defined. This review focuses on the roles of the various forms of endoglin in atherosclerosis. The expression of the two isoforms of endoglin (long-form and short-form) is increased in atherosclerotic lesions, and the expression of the soluble forms of endoglin is upregulated in sera of patients with hypercholesterolemia and atherosclerosis. Interestingly, long-form endoglin shows an atheroprotective effect via the induction of eNOS expression, while short-form and soluble endoglin enhance atherogenesis by inhibiting eNOS expression and TGF-β signaling. This review summarizes evidence suggesting that the different forms of endoglin have distinct roles in atherosclerosis.

[Immune Network 2014;14(5):237-240]

Keywords: Endoglin, Atherosclerosis, TGF-β, Smad

INTRODUCTION

TGF-β is a multi-functional cytokine and its signaling pathway contributes to a wide range of immunological and biological effects on various cell types and several diseases, TGF-β has regulatory functions in cell proliferation, differentiation, migration, and survival that affect multiple biological processes such as cell development, carcinogenesis, fibrosis, and wound healing, as well as immune responses (1). There is some controversy in the literature as to the precise functions of TGF-β; for example, in atherosclerosis, it is not clear whether TGF-β is pro-atherogenic or anti-atherogenic. The inhibition of TGF-β1 activity induces pro-atherogenic changes in the vessel wall of atherosclerotic animal models (2). Furthermore, the neutralization of TGF-β1 leads to an inflammatory response of the vessel wall and provokes plaque instability (3). After the engagement of TGF-β type I & II receptors, the Smad-dependent pathways are activated, The biological effects that result depend on the types of Smad complex involved in this signaling response (4). This process is modulated by other accessory receptors such as TGF-β type III receptor (5). The major type III receptor expressed in atherosclerotic lesions is endoglin. Although the expression of endoglin is increased in atherosclerotic lesions, the functional roles of endoglin in atherosclerosis have not been fully clarified. This review aims to suggest a hypothetical role for endoglin in atherosclerosis on the basis of previous reports.

STRUCTURE AND EXPRESSION

Endoglin (CD105) is a homodimer composed of two identical 95 kDa disulfide-linked subunits, and it is known as a hypoxia-inducible transmembrane glycoprotein (6). It consists of three domains: a large extracellular domain, a transmembrane domain, and a short intracellular domain. The extracellular domain contains an Arg-Gly-Asp (RGD) tri-peptide,
Endoglin was originally identified as a non-signaling coreceptor for TGF-β since it does not contain intrinsic kinase activity. The main function of endoglin is thought to be the regulation of TGF-β signaling via interactions with several proteins within the TGF-β signaling pathway. Endoglin binds to both of the TGF-β1 and TGF-β3 isoforms, following which the cytosolic domain of endoglin can be targeted by serine and threonine kinases, leading to the formation of a functional receptor complex (19). Indeed, endoglin is not a true receptor for TGF-β, but it strongly modulates the phosphorylation levels of TGF-βRII, activin receptor-like kinase (ALK)-1, and ALK-5 (20). The presence of endoglin can also modulate the downstream signaling by TGF-βRI/TGF-βRII complexes. Recent studies have demonstrated that endoglin functions in an interplay between two signaling pathways involving ALK-1 and ALK-5, respectively, that have differential effects on target cells. Endoglin/ALK-1/Smad1/5 signaling stimulates the migration, proliferation, and tube formation of endothelial cells, resulting in angiogenesis (21). In contrast, the endoglin/ALK-5/Smad2 pathway inhibits the activity of endothelial cells and angiogenesis by inhibiting the proliferation, tube formation, and migration of endothelial cells (22). In addition, endoglin has inhibitory effects on Smad3-dependent TGF-β signaling, resulting in effects on endothelial cells opposite to those that result from Smad2-dependent signaling (23). Mutations in endoglin have been reported in hereditary hemorrhagic telangiectasia, a disease characterized by malformations of vascular structure (24). The long-form and short-form endoglin isoforms are both able to bind to their ligands and interact with ALK-1 and ALK-5; however, the two membrane-bound endoglin isoforms differ in their affinity for each receptor, level of phosphorylation, and capacity to regulate TGF-β-dependent responses (25). Long-form endoglin has pro-angiogenic effects through induction of endogenous nitric oxide synthase (eNOS) expression, whereas short-form endoglin has anti-angiogenic effects. Thus, short-form endoglin contributes to the cardiovascular pathology associated with senescence. In addition, soluble endoglin, which inhibits TGF-β signaling, is thought to be cleaved from the cell membrane and enter the systemic circulation, and may represent a useful candidate marker of endothelial injury, activation, inflammation, and senescence (12).
ENDOGLIN AND ATHEROSCLEROSIS

Atherosclerosis is a chronic inflammatory disease of the arteries. Endothelial dysfunction plays an important role in the development of atherosclerosis by inducing infiltration of inflammatory cells and a prothrombogenic state. In addition, the migration and proliferation of smooth muscle cells, which are processes that affect the plaque stability, are crucial in the progression of advanced atherosclerotic lesions. TGF-β signaling results in inhibition of the proliferation and migration of smooth muscle cells, as well as endothelial cell regeneration (26). It was also reported that inhibition of TGF-β signaling reduced collagen content and plaque stability in a mouse model of atherosclerosis (3). In this model, TGF-β showed a protective effect against atherogenesis. Therefore, as an accessory receptor for TGF-β, endoglin expressed by endothelial cells and smooth muscle cells may play an important role in modifying the development of atherosclerosis via the regulation of TGF-β-induced atheroprotective effects. Although the expression of endoglin was very low in non-atherosclerotic aortas, the expression of endoglin by macrophages, smooth muscle cells and endothelial cells was increased in early atherosclerotic lesions (14). In advanced atherosclerotic plaques, smooth muscle cells expressed high levels of endoglin, a parameter that was independent of smooth muscle cell morphology and leukocyte infiltration. Moreover, endoglin modulates the expression of genes that are known to be related to pro-angiogenic effects (VEGF, angiopoietin-1, and angiopoietin-2) or anti-angiogenic effects (Notch signaling, Notch-3, and DLL4), respectively (27,28). Several studies have also reported that the concentration of soluble endoglin increased in the blood of patients with hypercholesterolemia and atherosclerosis (29). Increased soluble endoglin levels could be related to endothelial damage or dysfunction; furthermore, soluble endoglin is an indicator of cardiovascular damage in hypertension and diabetes-associated vascular pathologies (30). The circulating concentration of soluble endoglin was reported to increase at early stages of atherosclerosis due to damage of endothelial cells and then decrease in later stages of atherosclerosis, which suggests a potential role of soluble endoglin in acute heart failure (31). As a decoy receptor, soluble endoglin may inhibit the anti-atherogenic effects induced by TGF-β.

CONCLUSION

The expression levels of endoglin and soluble endoglin were higher in atherogenic lesions than in healthy arteries. Two isoforms of endoglin differentially transduce TGF-β signaling by activation of different Smad components, and thereby modulate the biological effects of TGF-β signaling. Long-form endoglin shows atheroprotective effect by induction of eNOS expression, while short-form endoglin and soluble endoglin forms enhance atherogenesis via down-regulation of eNOS expression and inhibition of TGF-β signaling.

ACKNOWLEDGEMENTS

This work was supported by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MEST) (NO. 2012R1A4A1029061).

CONFLICTS OF INTEREST

The authors have no financial conflict of interest.

REFERENCES

1. Massague, J., and D. Wotton. 2000. Transcriptional control by the TGF-beta/Smad signaling system. EMBO J. 19: 1745-1754.
2. Singh, N. N., and D. P. Ramji. 2006. The role of transforming growth factor-beta in atherosclerosis. Cytochrome Growth Factor Rev. 17: 487-499.
3. Mallat, Z., A. Gojova, C. Marchiol-Fournigault, B. Esposito, C. Karnate, R. Merval, D. Fradelizi, and A. Tedgui. 2001. Inhibition of transforming growth factor-beta signaling accelerates atherosclerosis and induces an unstable plaque phenotype in mice. Clin. Rev. 89: 930-934.
4. Feinberg, M. W., and M. K. Jain. 2005. Role of transforming growth factor-beta in atherosclerosis. Arterioscler. Thromb. Vasc. Biol. 25: 1745-1754.
6. Cheifetz, S., T. Bellon, C. Cales, S. Vera, C. Bernabeu, J. Massague, and M. Letarte. 1992. Endoglin is a component of the transforming growth factor-beta receptor system in human endothelial cells. J. Biol. Chem. 267: 19027-19030.
7. Gougos, A., and M. Letarte. 1990. Primary structure of endoglin, an RGD-containing glycoprotein of human endothelial cells. J. Biol. Chem. 265: 8361-8364.
8. Koleva, R. I., B. A. Conley, D. Romero, K. S. Riley, J. A. Marto, A. Lux, and C. P. Vary. 2006. Endoglin structure and...
function: Determinants of endoglin phosphorylation by transforming growth factor-beta receptors. *J. Biol. Chem.* 281: 25110-25123.

9. Fernandez-Ruiz, E., S. St-Jacques, T. Bellon, M. Letarte, and C. Bernabeu, 1993. Assignment of the human endoglin gene (END) to 9q34-qter. *Cytogenet. Cell Genet.* 64: 204-207.

10. St-Jacques, S., U. Gymerman, N. Pece, and M. Letarte, 1994. Molecular characterization and in situ localization of murine endoglin reveal that it is a transforming growth factor-beta binding protein of endothelial and stromal cells. *Endocrinology* 134: 2645-2657.

11. Bellon, T., A. Corbi, P. Lastres, C. Grelia, S. Vera, S. Cheltejz, J. Massague, M. Letarte, and C. Bernabeu, 1993. Identification and expression of two forms of the human transforming growth factor-beta binding protein endoglin with distinct cytoplasmic regions. *Eur. J. Immunol.* 23: 2340-2345.

12. Venkatash, S., M. Toporsian, C. Lam, J. Harai, T. Mamamoto, Y. M. Kim, Y. Boloh, K. H. Lim, H. T. Yuan, T. A. Libermann, I. E. Stillman, D. Roberts, P. A. D’Amore, F. H. Epstein, F. W. Sellke, R. Romero, V. P. Suhutme, M. Letarte, and S. A. Karumanchi. 2006. Soluble endoglin contributes to the pathogenesis of preclampsia. *Nat. Med.* 12: 642-649.

13. Hawinkels, L. J., P. Kuiper, E. Wiercinska, H. W. Verspaget, Z. Liu, E. Pardali, C. P. Vary. 2000. Endoglin, a TGF-beta receptor-associated protein, is expressed by smooth muscle cells in human atherosclerotic plaques. *Atherosclerosis* 153: 323-335.

14. Sanchez-Elsner, T., L. Botella, B. Velasco, C. Langa, and C. Bernabeu, 2002. Endoglin expression is regulated by transcriptional cooperation between the hypoxia and transforming growth factor-beta pathways. *J. Biol. Chem.* 277: 43799-43808.

15. Sanchez-Elsner, T., L. Botella, B. Velasco, C. Langa, and C. Bernabeu, 2002. Endoglin expression is regulated by transcriptional cooperation between the hypoxia and transforming growth factor-beta pathways. *J. Biol. Chem.* 277: 2340-2345.

16. Lastres, P., A. Letamendia, H. Zhang, C. Rius, N. Almendro, Y. M. Kim, Y. Bdolah, K. H. Lim, H. T. Yuan, T. A. Libermann, I. E. Stillman, D. Roberts, P. A. D’Amore, F. H. Epstein, F. W. Sellke, R. Romero, V. P. Suhutme, M. Letarte, and S. A. Karumanchi. 2006. Soluble endoglin contributes to the pathogenesis of preclampsia. *Nat. Med.* 12: 642-649.

17. Li, C., G. Pasterkamp, and A. Vink. 2008. Endoglin, a TGF-beta receptor-associated protein, is expressed by smooth muscle cells in human atherosclerotic plaques. *Atherosclerosis* 153: 323-335.

18. Sanchez-Elsner, T., L. Botella, B. Velasco, C. Langa, and C. Bernabeu, 2002. Endoglin expression is regulated by transcriptional cooperation between the hypoxia and transforming growth factor-beta pathways. *J. Biol. Chem.* 277: 43799-43808.

19. Santilanez, J. F., M. Quintanilla, and C. Bernabeu, 2011. TGF-beta/TGF-beta receptor system and its role in physiological and pathological conditions. *Clin. Sci. (Lond.)* 121: 233-251.

20. Guerrero-Esteo, M., T. Sanchez-Elsner, A. Letamendia, and C. Bernabeu, 2002. Extracellular and cytoplasmic domains of endoglin interact with the transforming growth factor-beta receptors I and II. *J. Biol. Chem.* 277: 29197-29209.

21. Ten Dijke, P., M. J. Goumans, and E. Pardali, 2008. Endoglin in angiogenesis and vascular diseases. *Angiogenesis* 11: 79-89.

22. Tian, F., A. X. Zhou, A. M. Smits, E. Larsson, M. J. Goumans, C. H. Heldin, J. Boren, and I. M. Akyurek. 2010. Endothelial cells are activated during hypoxia via endoglin/ALK-1/SMAD1/5 signaling in vivo and in vitro. *Biochem. Biophys. Res. Commun.* 392: 283-288.

23. Blanco, F. J., J. F. Santillan, M. Guerrero-Esteo, C. Langa, C. P. Vary, and C. Bernabeu, 2005. Interaction and functional interplay between endoglin and ALK-1, two components of the endothelial transforming growth factor-beta receptor complex. *J. Cell Physiol.* 204: 574-584.

24. Llorca, O., A. Trajillo, F. J. Blanco, and C. Bernabeu, 2007. Structural model of human endoglin, a transmembrane receptor responsible for hereditary hemorrhagic telangiectasia. *J. Mol. Biol.* 365: 694-705.

25. Blanco, F. J., M. T. Grande, C. Langa, B. Ojo, S. Velasco, A. Rodriguez-Barbero, E. Perez-Gomez, M. Quintanilla, J. M. Lopez-Navoa, and C. Bernabeu, 2008. sEndoglin expression is induced in senescent endothelial cells and contributes to vascular pathology. *Circ. Res.* 103: 1383-1392.

26. Gamble, J. R., Y. Khew-Goodall, and M. A. Vadas. 1993. Transforming growth factor-beta inhibits E-selectin expression on human endothelial cells. *J. Immunol.* 150: 4494-4503.

27. Li, C., P. Mollahan, M. S. Baguneid, R. F. McMahon, P. Kumar, M. G. Walker, A. J. Freemont, and S. Kumar, 2006. A comparative study of neovascularisation in atherosclerotic plaques using CD31, CD105 and TGF beta 1. *Pathobiology* 73: 192-197.

28. Post, S., W. Peeters, E. Busser, D. Lamers, J. P. Sluijter, M. J. Goumans, R. A. de Weger, F. L. Moli, P. A. Dövenlands, G. Pasterkamp, and A. Vink. 2008. Balance between angiopeptin-1 and angiopeptin-2 is in favor of angiopeptin-2 in atherosclerotic plaques with high microvessel density. *J. Vasc. Res.* 45: 244-250.

29. Blann, A. D., J. M. Wang, P. B. Wilson, and S. Kumar, 1996. Serum levels of the TGF-beta receptor are increased in atherosclerosis. *Atherosclerosis* 120: 221-228.

30. Blazquez-Medela, A. M., L. Garcia-Ortiz, A. Sanchez-Rodriguez, M. J. Goumans, R. A. de Weger, F. L. Moli, P. A. Dövenlands, G. Pasterkamp, and A. Vink. 2008. Balance between angiopeptin-1 and angiopeptin-2 is in favor of angiopeptin-2 in atherosclerotic plaques with high microvessel density. *J. Vasc. Res.* 45: 244-250.