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# Disabled-2 Is Essential for Endodermal Cell Positioning and Structure Formation during Mouse Embryogenesis ☆

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## Abstract

The signal transduction adapter protein Disabled-2 (Dab2) is one of the two mammalian orthologs of the *Drosophila* Disabled. The brain-specific Disabled-1 (Dab1) functions in positional organization of brain cells during development. Dab2 is widely distributed and is highly expressed in many epithelial cell types. The *dab2* gene was interrupted by in-frame insertion of β-galactosidase (LacZ) in embryonic stem cells and transgenic mice were produced. Dab2 expression was first observed in the primitive endoderm at E4.5, immediately following implantation. The homozygous Dab2-deficient mutant is embryonic lethal (earlier than E6.5) due to defective cell positioning and structure formation of the visceral endoderm. In E5.5 *dab2* (−/−) conceptus, visceral endoderm-like cells are present in the deformed primitive egg cylinder; however, the visceral endoderm cells are not organized, the cells of the epiblast have not expanded, and the proamniotic cavity fails to form. Disorganization of the visceral endodermal layer is evident, as cells with positive visceral endoderm markers are scattered throughout the *dab2* (−/−) conceptus. Only degenerated remains were observed at E6.5 for *dab2* (−/−) embryos, and by E7.5, the defective embryos were completely reabsorbed. In blastocyst *in vitro* culture, initially cells with characteristics of endoderm, trophectoderm, and inner cell mass were observed in the outgrowth of the hatched *dab2* (−/−) blastocysts. However, the *dab2* (−/−) endodermal cells are much more dispersed and disorganized than those from wild-type blastocysts, the inner cell mass fails to expand, and the outgrowth degenerates by day 7. Thus, Dab2 is required for visceral endodermal cell organization during early mouse development. The absence of an organized visceral endoderm in Dab2-deficient conceptus leads to the growth failure of the inner cell mass. We suggest that Dab2 functions in a signal pathway to regulate endodermal cell organization using endocytosis of ligands from the blastocoel cavity as a positioning cue.

## Keywords

Disabled-2; visceral endoderm; cell positioning/organization; embryos; extraembryonic endoderm; blastocysts; differentiation; endocytosis



## References

REFERENCES

1 R.J. Arceci, A.A. King, M.C. Simon, S.H. Orkin, D.B. Wilson
**Mouse GATA-4: A retinoic acid-inducible GATA-binding transcription factor expressed in endodermally derived tissues and heart**
Mol. Cell. Biol., 13 (1993), pp. 2235-2246

2 E. Barbacci, M. Reber, M.O. Ott, C. Breillat, F. Huetz, S. Cereghini
**Variant hepatocyte nuclear factor 1 is required for visceral endoderm specification**
Development, 126 (1999), pp. 4795-4805

3 M. Bielinska, N. Narita, D.B. Wilson
**Distinct roles for visceral endoderm during embryonic mouse development**
Int. J. Dev. Biol., 43 (1999), pp. 183-205

4 T. Burdon, C. Stracey, I. Chambers, J. Nichols, A. Smith
**Suppression of SHP-2 and ERK signalling promotes self-renewal of mouse embryonic stem cells**
Dev. Biol., 210 (1999), pp. 30-43
Article PDF (499KB)

5 W.S. Chen, K. Manova, D.C. Weinstein, S.A. Duncan, A.S. Plump, V.R. Prezioso, R.F. Bachvarova, J.E. Darnell Jr.
**Disruption of the HNF-4 gene, expressed in visceral endoderm, leads to cell death in embryonic ectoderm and impaired gastrulation of mouse embryos**
Genes Dev., 8 (1994), pp. 2466-2477

6 A.M. Cheng, T. Saxton, M. Sakai, R.S. Kulkarni, G. Mbamalu, W. Vogel, C.G. Tortorice, R.D. Cardiff, J.C. Cross, W.J. Muller, T. Pawson
**Mammalian Grb2 regulates multiple steps in embryonic development and malignant transformation**
Cell, 95 (1998), pp. 793-803
Article PDF (575KB)

7 C. Coffinier, D. Thepot, C. Babinet, M. Yaniv, J. Barra
**Essential role for the homeoprotein vHNF1/HNF1beta in visceral endoderm differentiation**
Development, 126 (1999), pp. 4785-4794

8 E. Coucouvanis, G.R. Martin
**Signals for death and survival: A two-step mechanism for cavitation in the vertebrate embryo**
Cell, 83 (1995), pp. 279-287
Article PDF (14MB)

9 E. Coucouvanis, G.R. Martin
**BMP signaling plays a role in visceral endoderm differentiation and cavitation in the early mouse embryo**
Development, 126 (1999), pp. 535-546

10 G. D'Arcangelo, G.G. Miao, S.C. Chen, H.D. Soares, J.I. Morgan, T. Curran
**A protein related to extracellular matrix proteins deleted in the mouse mutant reeler**
Nature, 374 (1995), pp. 719-723

11 G. D'Arcangelo, K. Nakajima, T. Miyata, M. Ogawa, K. Mikoshiba, T. Curran
**Reelin is a secreted glycoprotein recognized by the CR-50 monoclonal antibody**
J. Neurosci., 17 (1997), pp. 23-31

12 M. Dziadek
**Modulation of alphafetoprotein synthesis in the early postimplantation mouse embryo**
J. Embryol. Exp. Morphol., 46 (1978), pp. 135-146

13 R.V. Farese Jr., S.L. Ruland, L.M. Flynn, R.P. Stokowski, S.G. Young
**Knockout of the mouse apolipoprotein B gene results in embryonic lethality in homozygotes and protection against diet-induced hypercholesterolemia in heterozygotes**
Proc. Natl. Acad. Sci. USA, 92 (1995), pp. 1774-1778

14 R.V. Farese Jr., J. Herz
**Cholesterol metabolism and embryogenesis**
Trends Genet., 14 (1998), pp. 115-120

15 Z. Fazili, W. Sun, S. Mittelstaedt, C. Cohen, X.X. Xu
**Disabled-2 inactivation is an early step in ovarian tumorigenicity**
Oncogene, 18 (1999), pp. 3104-3113

Oncogene, 18 (1999), pp. 6104-6113

16 C. Gueth-Hallonet, A. Santa-Maria, P. Verroust, B. Maro
**Gp330 is specifically expressed in outer cells during epithelial differentiation in the preimplantation mouse embryo**
Development, 120 (1994), pp. 3289-3299

17 J. He, E.R. Smith, X.X. Xu
**Disabled-2 exerts its tumor suppressor activity by uncoupling c-Fos expression and MAP kinase activation**
J. Biol. Chem., 276 (2001), pp. 26814-26818

18 J. Herz, D.E. Clouthier, R.E. Hammer
**LDL receptor-related protein internalizes and degrades uPA-PAI-1 complexes and is essential for embryo implantation**
Cell, 71 (1992), pp. 411-421
Article PDF (1MB)

19 J. Herz, D.E. Clouthier, R.E. Hammer
**Erratum in: LDL receptor-related protein internalizes and degrades uPA-PAI-1 complexes and is essential for embryo implantation**
Cell, 73 (1993), p. 428
Article PDF (70KB)

20 J. Herz, R.V. Farese Jr.
**The LDL receptor gene family, apolipoprotein B and cholesterol in embryonic development**
J. Nutr., 129 (1999), pp. 473S-475S

21 B.A. Hocevar, A. Smine, X.X. Xu, P.H. Howe
**The adaptor molecule Disabled-2 links the transforming growth factor beta receptors to the Smad pathway**
EMBO J., 20 (2001), pp. 2789-2801

22 F.M. Hoffmann
**Drosophila abl and genetic redundancy in signal transduction**
Trends Genet., 7 (1991), pp. 351-355
Article PDF (643KB)

23 B.L. Hogan, A. Taylor, E. Adamson
**Cell interactions modulate embryonal carcinoma cell differentiation into parietal or visceral endoderm**
Nature, 291 (1981), pp. 235-237

24 B.L. Hogan, R. Tilly
**Cell interactions and endoderm differentiation in cultured mouse embryos**
J. Embryol. Exp. Morphol., 62 (1981), pp. 379-394

25 B.L. Hogan, R. Newman
**A scanning electron microscope study of the extraembryonic endoderm of the 8th-day mouse embryo**
Differentiation, 26 (1984), pp. 138-143
Article PDF (1MB)

26 B.W. Howell, F.B. Gertler, J.A. Cooper
**Mouse disabled (mDab1): A Src binding protein implicated in neuronal development**
EMBO J., 16 (1997), pp. 121-132

27 B.W. Howell, R. Hawkes, P. Soriano, J.A. Cooper
**Neuronal position in the developing brain is regulated by mouse disabled-1**
Nature, 389 (1997), pp. 733-737

28 B.W. Howell, L. Lanier, M.R. Frank, F.B. Gertler, J.A. Cooper
**The disabled 1 phosphotyrosine-binding domain binds to the internalization signals of transmembrane glycoproteins and to phospholipids**
Mol. Cell. Biol., 19 (1999), pp. 5179-5188

29 B.W. Howell, J. Herz
**The LDL receptor gene family: Signaling functions during development**
Curr. Opin. Neurobiol., 11 (2001), pp. 74-81
Article PDF (111KB)

30 L.S. Huang, E. Voyiaziakis, D.F. Markenson, K.A. Sokol, T. Hayek, J.L. Breslow
**apo B gene knockout in mice results in embryonic lethality in homozygotes and neural tube defects, male infertility, and reduced HDL cholesterol ester and apo A-I transport rates in heterozygotes**
J. Clin. Invest., 96 (1995), pp. 2152-2161

31 M.Z. Kounnas, C.C. Haudenschild, D.K. Strickland, W.S. Argraves
**Immunological localization of glycoprotein 330, low density lipoprotein receptor related protein and 39 kDa receptor associated protein in embryonic mouse tissues**
In Vivo, 8 (1994), pp. 343-351

32 M. Koutsourakis, A. Langeveld, R. Patient, R. Beddington, F. Grosveld
**The transcription factor GATA6 is essential for early extraembryonic development**
Development, 126 (1999), pp. 723-732

33 C.C. Lu, J. Brennan, E.J. Robertson
**From fertilization to gastrulation: Axis formation in the mouse embryo**
Curr. Opin. Genet. Dev., 11 (2001), pp. 384-392
Article PDF (271KB)

34 P. Maye, S. Becker, E. Kasameyer, N. Byrd, L. Grabel
**Indian hedgehog signaling in extraembryonic endoderm and ectoderm differentiation in ES embryoid bodies**
Mech. Dev., 94 (2000), pp. 117-132
Article PDF (2MB)

35 S.C. Mok, W.Y. Chan, K.K. Wong, K.K. Cheung, C.C. Lau, S.W. Ng, A. Baldini, C.V. Colitti, C.O. Rock, R.S. Berkowitz
**DOC-2, a candidate tumor suppressor gene in human epithelial ovarian cancer**
Oncogene, 16 (1998), pp. 2381-2387

36 S.M. Morris, J.A. Cooper
**Disabled-2 colocalizes with the LDLR in clathrin-coated pits and interacts with AP-2**
Traffic, 2 (2001), pp. 111-123

37 S.M. Morris, M.D. Tallquist, C.O. Rock, J.A. Cooper
**Dual roles for the Dab2 adaptor protein in embryonic development and kidney transport**
EMBO J., 21 (2002), pp. 1555-1564

38 E.E. Morrisey, Z. Tang, K. Sigrist, M.M. Lu, F. Jiang, H.S. Ip, M.S. Parmacek
**GATA6 regulates HNF4 and is required for differentiation of visceral endoderm in the mouse embryo**
Genes Dev., 12 (1998), pp. 3579-3590

39 E.E. Morrisey, S. Musco, M.Y. Chen, M.M. Lu, J.M. Leiden, M.S. Parmacek
**The gene encoding the mitogen-responsive phosphoprotein Dab2 is differentially regulated by GATA-6 and GATA-4 in the visceral endoderm**
J. Biol. Chem., 275 (2000), pp. 19949-19954

40 A. Nose, M. Takeichi
**A novel cadherin cell adhesion molecule: its expression patterns associated with implantation and organogenesis of mouse embryos**
J. Cell Biol., 103 (1986), pp. 2649-2658

41 A.V. Oleinikov, J. Zhao, S.P. Makker
**Cytosolic adaptor protein Dab2 is an intracellular ligand of endocytic receptor gp600/megalin**
Biochem. J., 347 (2000), pp. 613-621

42 D.S. Rice, T. Curran
**Mutant mice with scrambled brains: Understanding the signaling pathways that control cell positioning in the CNS**
Genes Dev., 13 (1999), pp. 2758-2773

43 M. Sato, T. Muramatsu
**Reactivity of five N-acetylgalactosamine-recognizing lectins with preimplantation embryos, early postimplantation embryos, and teratocarcinama cells of the mouse**
Differentiation, 29 (1985), pp. 29-38

Article PDF (1MB)

44 M. Sheldon, D.S. Rice, G. D'Arcangelo, H. Yoneshima, K. Nakajima, K. Mikoshiba, B.W. Howell, J.A. Cooper, D. Goldowitz, T. Curran
**Scrambler and Yotari disrupt the disabled gene and produce a reeler-like phenotype in mice**
Nature, 389 (1997), pp. 730-733

45 Z. Sheng, W. Sun, E.R. Smith, C. Cohen, Z. Sheng, X.X. Xu
**Restoration of positioning control following Disabled-2 expression in ovarian and breast tumor cells**
Oncogene, 19 (2000), pp. 4847-4854

46 Z. Sheng, E.R. Smith, J.A. Tuppen, W.D. Martin, F.B. Dong, X.X. Xu
**Chromosomal location of murine Disabled-2 and structural comparison with its human ortholog**
Gene, 268 (2001), pp. 31-39
Article PDF (589KB)

47 M.I. Sherman, R.A. Miller
**F9 embryonal carcinoma cells can differentiate into endoderm-like cells**
Dev. Biol., 63 (1978), pp. 27-34
Article PDF (3MB)

48 E.R. Smith, J.L. Smedberg, M.E. Rula, T.C. Hamilton, X.X. Xu
**Disassociation of MAPK activation and c-Fos expression in F9 embryonic carcinoma cells following retinoic acid-induced endoderm differentiation**
J. Biol. Chem., 276 (2001), pp. 32094-32100

49 E.R. Smith, C.D. Capo-chichi, J. He, J.L. Smedberg, M.E. Rula, D.H. Yang, A.H. Prowse, A.K. Godwin, T.C. Hamilton, X.X. Xu
**Disabled-2 mediates c-Fos suppression and the cell growth regulatory activity of retinoic acid in embryonic carcinoma cells**
J. Biol. Chem., 276 (2001), pp. 47303-47310

50 N. Smyth, H.S. Vatansever, P. Murray, M. Meyer, C. Frie, M. Paulsson, D. Edgar
**Absence of basement membranes after targeting the LAMC1 gene results in embryonic lethality due to failure of endoderm differentiation**
J. Cell Biol., 144 (1999), pp. 151-160

51 C. Soudais, M. Bielinska, M. Heikinheimo, C.A. MacArthur, N. Narita, J.E. Saffitz, M.C. Simon, J.M. Leiden, D.B. Wilson
**Targeted mutagenesis of the transcription factor GATA-4 gene in mouse embryonic stem cells disrupts visceral endoderm differentiation in vitro**
Development, 121 (1995), pp. 3877-3888

52 D.D. Spyropoulos, M.R. Capecchi
**Targeted disruption of the even-skipped gene, evx1, causes early postimplantation lethality of the mouse conceptus**
Genes Dev., 8 (1994), pp. 1949-1961

53 Y. Terasawa, S.J. Cases, J.S. Wong, H. Jamil, S. Jothi, M.G. Traber, L. Packer, D.A. Gordon, R.L. Hamilton, R.V. Farese Jr.
**Apolipoprotein B-related gene expression and ultrastructural characteristics of lipoprotein secretion in mouse yolk sac during embryonic development**
J. Lipid Res., 40 (1999), pp. 1967-1977

54 M. Trommsdorff, J.P. Borg, B. Margolis, J. Herz
**Interaction of cytosolic adaptor proteins with neuronal apolipoprotein E receptors and the amyloid precursor protein**
J. Biol. Chem., 273 (1998), pp. 33556-33560

55 M. Trommsdorff, M. Gotthardt, T. Hiesberger, J. Shelton, W. Stockinger, J. Nimpf, R.E. Hammer, J.A. Richardson, J. Herz
**Reeler/Disabled-like disruption of neuronal migration in knockout mice lacking the VLDL receptor and ApoE receptor 2**
Cell, 97 (1999), pp. 689-701
Article PDF (1MB)

56 M.H. Verheijen, R.M. Wolthuis, J.L. Bos, L.H. Defize
**The Ras/Erk pathway induces primitive endoderm but prevents parietal endoderm differentiation of F9 embryonal carcinoma cells**
J. Biol. Chem., 274 (1999), pp. 1487-1494

57 T.E. Willnow, J. Hilpert, S.A. Armstrong, A. Rohlmann, R.E. Hammer, D.K. Burns, J. Herz
**Defective forebrain development in mice lacking gp330/megalin**

**Defective forebrain development in mice lacking gp330/megalin**
Proc. Natl. Acad. Sci. USA, 93 (1996), pp. 8460-8464

58 T.E. Willnow, A. Nykjaer, J. Herz
**Lipoprotein receptors: New roles for ancient proteins**
Nat. Cell Biol., 1 (1999), pp. E157-E162

59 X.X. Xu, W. Yang, S. Jackowski, C.O. Rock
**Molecular cloning of a CSF-1 regulated protein with a domain homologous to Drosophila disabled**
J. Biol. Chem., 270 (1995), pp. 14184-14191

60 D.H. Yang, S. Tsuyama, Y.B. Ge, D. Wakamatsu, J. Ohmori, F. Murata
**Proliferation and migration kinetics of stem cells in the rat fundic gland**
Histol. Histopathol., 12 (1997), pp. 719-727

61 D.H. Yang, E.R. Smith, C. Cohen, C. Patriotis, A.K. Godwin, T.C. Hamilton, X.X. Xu
**Molecular events associated with dysplastic morphological transformation and initiation of ovarian tumorigenicity**
Cancer, 94 (2002), pp. 2380-2392

☆ Abbreviations used: AFP, alpha fetal protein; ApoE2R, Apolipoprotein E2 receptor; BM, basement membrane; BMP, bone morphogenesis protein; Dab2, Disabled-2 (protein); DAB2, human Disabled-2 gene; *dab2*, mouse Disabled-2 gene; DBA, *Dolichos biflorus* agglutinin; E, embryonic day; EC, embryonic carcinoma; ES cells, embryonic stem cells; evx1, even-skipped gene 1; FBS, fetal bovine serum; Grb2, growth factor receptor binding protein 2; HNF, hepatocyte nuclear factor; Ihh, Indian hedgehog; ICM, inner cell mass; LDLR, low density lipoprotein receptor; LIF, leukemia inhibitory factor; LRP, low density lipoprotein receptor related protein; MAPK (Erk), mitogen-activated protein kinase (Erk, extracellular-signal regulated kinase); MEK, kinase for MAPK or Erk; PAS staining, periodic acid-Schiff staining; PE, parietal endoderm; PID/PTB, phosphotyrosine interacting domain or phosphotyrosine binding domain; Sos, Son-of-Sevenless; RA, retinoic acid; TdT, terminal dTTP transferase; VE, visceral endoderm; vHNF, variant hepatocyte nuclear factor; VLDLR, very low density lipoprotein receptor.

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