



**Fig. S1. No inclusion of *ASTL* expressing cells in isolated area pellucida or area opaca.** An inner part of area pellucida or an outer part of area opaca is excised from the pre-primitive-streak embryos and checked for the expression of the marginal zone marker *ASTL* after 8 h culture (as conducted in Fig. 1). (A) *ASTL* expression in a pre-primitive-streak embryo. (B and C) no expression of *ASTL* in the isolated area pellucida (B) or area opaca (C). (D) *ASTL* expression in the remaining tissue containing a bit of area opaca, marginal zone and area pellucida, after excision of the area opaca.