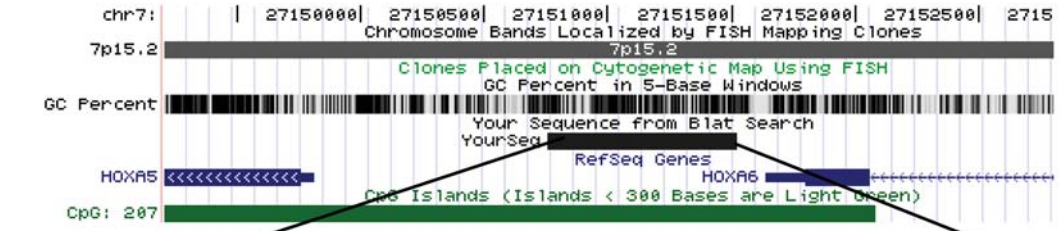
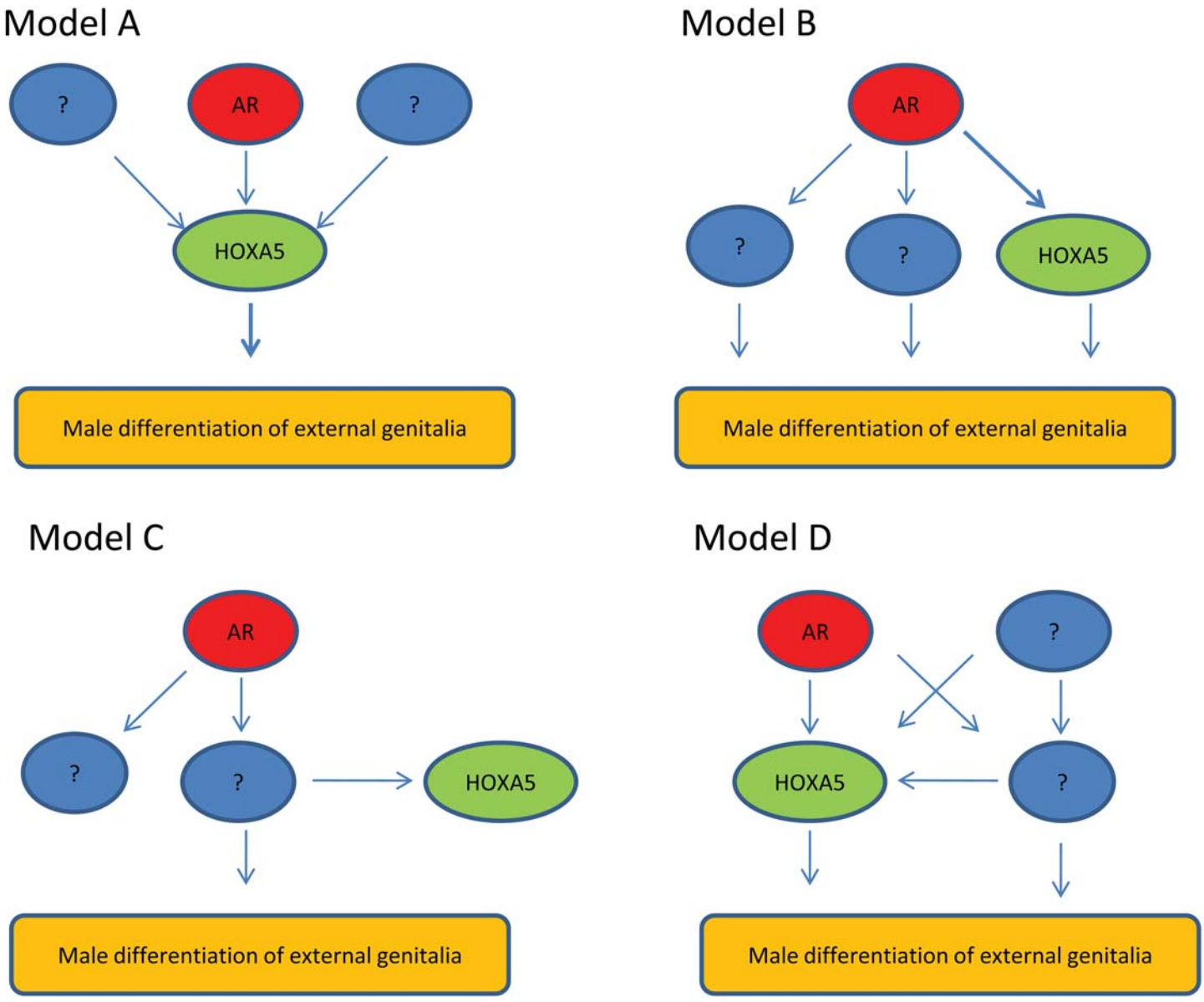


Phenotype/CpG	1	2	3	4	5	6	7	8	9	10	11	12	13	14	Genotype
CAIS	81	81	87	81	83	93	86	90	92	61	66	62	88	57	p.E472fs
CAIS	68	69	70	71	74	76	72	76	52	57	76	48	75	64	p.R855C
CAIS	17	12	15	13	14	17	18	16	10	9	15	10	15	13	p.P390S+R855G
CAIS	41	44	47	39	24	47	59	39	23	29	26	34	27	24	p.E287X
CAIS	61	51	54	53	67	71	72	43	30	38	36	41	60	46	p.Q59X
AIS4	31	21	13	17	27	36	27	22	10	7	19	14	15	17	p.I841S
AIS4	29	26	17	16	18	33	31	19	12	9	17	11	17	10	p.A870G
AIS3	81	46	45	44	45	60	83	47	44	42	55	61	87	33	p.=(c.3026C>T)
AIS3	47	24	25	17	26	29	24	18	11	11	38	16	72	27	p.R608K
AIS2	43	37	38	38	51	54	57	45	28	25	39	37	41	33	p.=(c.2747A>G)
AIS2	76	71	71	66	72	77	74	75	60	43	71	55	69	58	p.L712F
AIS2	30	23	23	20	69	75	76	34	25	15	67	21	32	30	p.A645D
AIS2	38	20	24	28	32	33	33	26	19	17	26	16	29	23	p.R855H
AIS2	68	53	56	44	83	74	59	60	21	49	58	39	53	53	p.A596Y
MAIS	83	79	82	83	84	89	84	94	92	75	85	63	89	72	p.I841S
control 1	13	12	14	11	77	87	20	17	12	10	16	40	20	70	
control 2	48	11	11	12	42	16	48	14	12	7	50	8	28	42	
control 3	18	12	19	16	12	22	20	16	12	16	20	11	16	11	
control 4	45	32	12	9	43	56	60	38	12	7	34	32	63	14	
control 5	32	15	20	24	38	35	47	32	19	10	42	22	53	27	
control 6	33	24	26	22	33	36	36	29	17	14	34	23	28	16	
control 7	36	23	30	25	25	32	38	37	19	20	27	26	33	20	
control 8	41	21	20	27	28	47	52	39	18	15	29	27	37	24	
control 9	40	29	29	26	29	38	54	30	19	18	35	21	29	21	
UM control	7	7	9	6	7	6	10	11	10	9	11	5	12	8	
M control	90	91	93	90	92	97	91	99	97	95	100	74	96	85	

Suppl. Fig. 1

The upper part of the figure depicts the location of the analyzed region on chromosome 7. This part of the figure was created with the UCSC Human Genome Browser Gateway. The table depicts the methylation raw data from pyrosequencing. It shows for each individual tested and each CpG position (1–14) a methylation value in %. Exel conditional formatting was used with setting 100% methylation as red, 50% methylation as black and 0% methylation as green to visualize methylation differences. For AIS patients the table shows on the left side the phenotype and on the right side the genotype. CAIS: complete androgen insensitivity syndrome, female external genitalia. AIS4: slight virilization, predominantly female external genitalia. AIS3: ambiguous external genitalia. AIS2: predominantly male external genitalia. MAIS: male external genitalia. UM control: unmethylated control sample, M control: methylated control sample.



Suppl. Fig. 2
Models A–D depict possible explanations for the early embryonic androgen pathway resulting in final male differentiation of the external genitalia. Model A depicts HOXA5 as an androgen receptor regulated gene. The fact that identical AR mutations can lead to different HOXA5 methylation patterns is addressed by the hypothesis of additional, independent gene influences on HOXA5. Model B was created in order to explain AIS due to AR mutation in the case of normal HOXA5 DNA methylation. Other androgen regulated genes may in these cases contribute to the AIS phenotype. Model C depicts the possibility that HOXA5 is only indirectly influenced by the AR; in that case HOXA5 would not directly contribute to final male differentiation. Model D integrates Model A and B and explains all our findings. It leaves lots of variables open for further research in the field of the AR pathway and its influence on AIS.