SUPPLEMENTARY MATERIAL

Mutations in the DNA methyltransferase, *DNMT3A*, cause a novel overgrowth syndrome

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Supplementary Table 1. *DNMT3A* primer sequences

Exon	Forward sequence	Reverse sequence	PCR product size (bp)	Multiplex group
Exon 2	ACCACGATAATTCCTTCCCC	CGGCTGTCATCACATAGGG	200	A
Exon 3	AGACAGGCCTGGAATGC	CCCTGCAGGACATACATCAC	263	B
Exon 4	AGCGGTCAATGATCCAAAAC	GGAGAGCCAAGTCCCTGAC	419	C
Exon 5	GGCCTGTGAGCCACTCC	TCACACACACTCGCCACC	185	D
Exon 6	CATTGTGTTTGAGGCGAGTG	GAGGCCCATCACTTCTGG	296	A
Exon 7A	ATGGTCCCCTTGAGTGTCAG	TGCTTTGGTGGCATTCTTG	269	B
Exon 7B	CCGGGGAGTCTCAGAAGG	GAGGAGCTGGCAGTGGAAG	221	D
Exon 8	CCCTCCTGGCTGTCCTG	CACTTCCATCACCCCAATTC	375	C
Exon 9	CTGACTCTCGAGGCTCCCAG	ACCTGCACTCCAACTTCCAG	277	Single PCR
Exon 10	CTCAGAGTCTGGCCTTGAGC	CTCCCTAAGCATGGCTTTCC	319	A
Exon 11	GGCTGGAGTTTCCTGTCAGC	GAGAGCTGGCGTCAGAGG	289	C
Exon 12	CACTGTAAGGAGGGTGGGG	AACCTTCCTAAGTGCCTCTGC	445	Single PCR
Exon 13	TTTTGTGGCTGGTCTGGTG	AAGCGGTGGACACAGTCAG	245	C
Exon 14	GTGGAGCCTCCCTTCGTC	GGAGGGTCTGTGGGGAAG	281	D
Exon 15	TCCATTCCAGGTAGCACACC	TCCTAGACCCACACACCCTG	352	A
Exon 16	GTGGGAGCTTGGGACACC	TTCGTTTTGCCAGAGTTGC	225	B
Exon 17	CTCACCTGCCGAGACCAG	ATGAAAGGAGGCAAGGGC	296	C
Exon 18	GACTTCTTTGCCAAGTTCACAG	GGCAGAAATATCCAAGGAGG	261	D
Exon 19	ATGACCCTGTCTTCCCGTG	GAGACAGGATGAAGCAGCAG	316	A
Exon 20	CTTTAAGGCTCGACCCCAG	CTTCCCCACTATGGGTCATC	240	B
Exon 21	TGTGAACTAGTGGCTGCTGG	CGCTCCACCTCATCCTG	240	C
Exon 22	CTGGCATATTTGGTAGACGC	AGTCAGGTGGGAAAGGCAG	289	D
Exon 23	TCCTGCTGTGTGGTTAGACG	AAACAAACAGAAAACCCCTCTG	552	A
Sequencing primer	**Primer sequence**	**Direction**		
Exon 3	ACACTGCTGGGATCCACCTC	Forward		
Exon 6	CTTCTGGTTTTCCAGTTCTGC	Reverse		
Exon 8	GGGCCTCGTGACCACTG	Forward		
Exon 10	ATCTGCCTTCCAGCCTGTC	Forward		
Exon 14	AGGAGGGGAGGCGGTG	Reverse		
Exon 22	CAGCAAGCACAGCAATCAG	Reverse		

Supplementary Table 2. Somatic *DNMT3A* mutations in haematological neoplasms

Mutation	Mutation type	Domain	Count
p.Gln110fs	Indel	None	1
p.Gln153X	Stop gain	None	1
p.Gln237X	Stop gain	None	1
p.Gly332fs	Indel	PWWP	1
p.Ser352Asn	nonsynonymous	None	1
p.Leu422fs	Indel	None	1
p.Lys468Arg	nonsynonymous	None	1
p.Glu477X	Stop gain	None	2
p.Glu505X	Stop gain	ADD	2
p.Gln515X	Stop gain	ADD	1
p.Ser535fs	Indel	ADD	2
p.Gly543Cys	nonsynonymous	ADD	1
p.Trp581Cys	nonsynonymous	ADD	1
p.Gly590fs	Indel	ADD	2
p.Glu599X	Stop gain	ADD	1
p.Gln615X	Stop gain	None	1
p.Arg635Trp	nonsynonymous	Mtase	1
p.Val636Met	nonsynonymous	Mtase	1
p.Ile655Asn	nonsynonymous	Mtase	1
p.Arg676Trp	nonsynonymous	Mtase	1
p.Gly685Arg	nonsynonymous	Mtase	1
p.Gly699Ser	nonsynonymous	Mtase	1
p.Ser714Cys	nonsynonymous	Mtase	1
p.Pro718Leu	nonsynonymous	Mtase	1
p.Arg720Cys	nonsynonymous	Mtase	1
p.Leu723fs	Indel	Mtase	3
p.Arg729Gln	nonsynonymous	Mtase	1
p.Arg729Trp	nonsynonymous	Mtase	1
p.Phe731del	Inframe deletion	Mtase	6
p.Phe731Cys	nonsynonymous	Mtase	1
p.Tyr735Cys	nonsynonymous	Mtase	1
p.Arg736His	nonsynonymous	Mtase	1
p.Leu737Arg	nonsynonymous	Mtase	1
p.Ala741Val	nonsynonymous	Mtase	2
p.Asp748Asn	nonsynonymous	Mtase	1
p.Arg749Cys*	nonsynonymous	Mtase	1
p.Phe751fs	Indel	Mtase	1
p.Phe752Leu	nonsynonymous	Mtase	2
p.Trp753Gly	nonsynonymous	Mtase	1
p.Leu754Arg	nonsynonymous	Mtase	1
p.Ala760fs	Indel	Mtase	1
p.Gly762ins2	inframe insertion	Mtase	4
p.Lys766fs	Indel	Mtase	1
p.Ser770Trp	nonsynonymous	Mtase	1
p.Arg771Leu	nonsynonymous	Mtase	1
p.Arg792His	nonsynonymous	Mtase	1
p.Trp795Cys	nonsynonymous	Mtase	2
p.Arg803Ser	nonsynonymous	Mtase	1
p.Lys829Arg	nonsynonymous	Mtase	1
p. Lys841Gln	nonsynonymous	Mtase	1

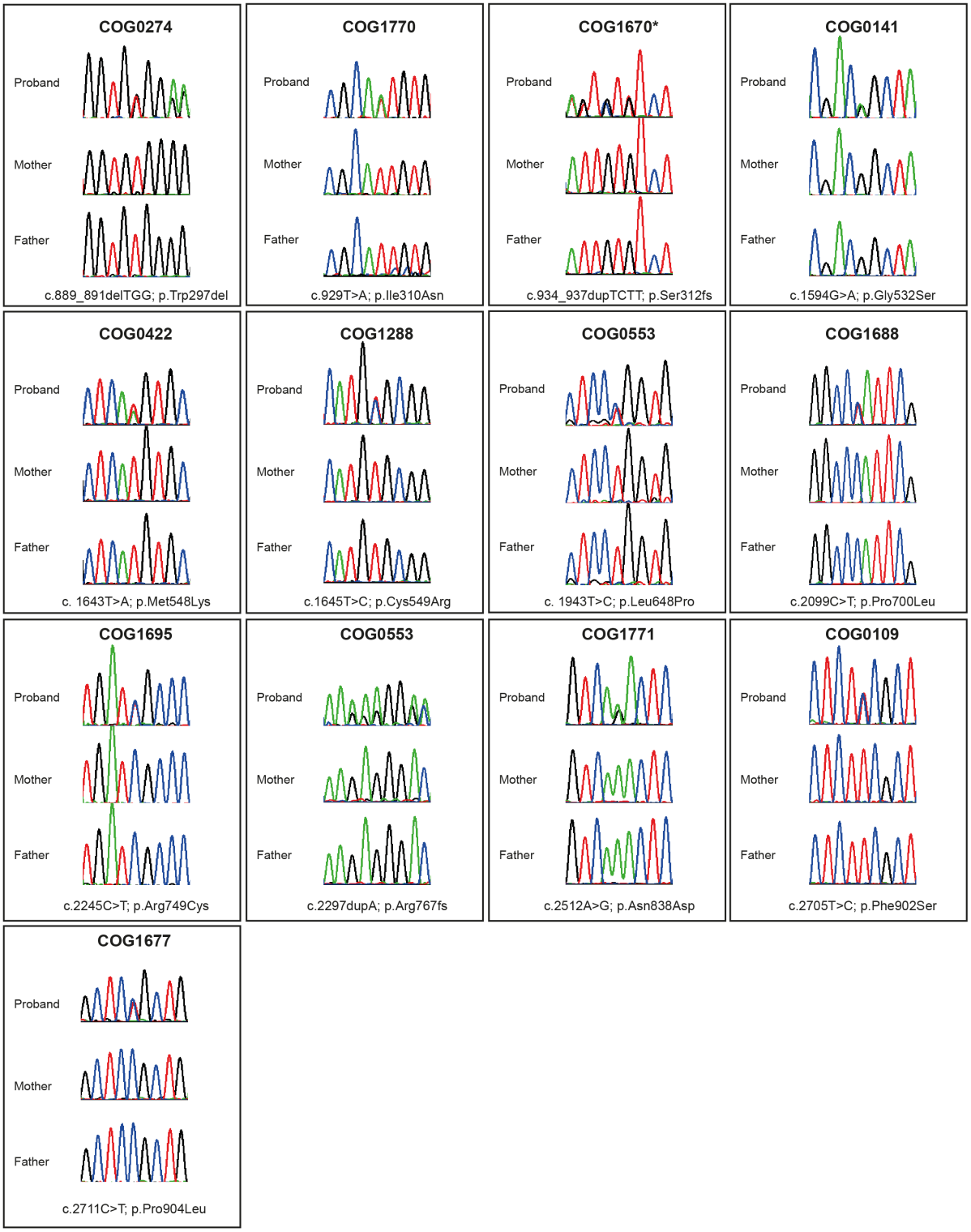
Mutation	Mutation type	Domain	Count
p.Pro849Arg	nonsynonymous	Mtase	1
p.Asp857fs	Indel	Mtase	1
p.Met880Val	nonsynonymous	Mtase	2
p.Arg882Cys	nonsynonymous	Mtase	23
p.Arg882His	nonsynonymous	Mtase	61
p.Arg882Pro	nonsynonymous	Mtase	1
p.Leu889Pro	nonsynonymous	Mtase	1
p.Val897Asp	nonsynonymous	Mtase	1
p.Phe902fs	Indel	Mtase	1
p.Pro904Leu*	nonsynonymous	Mtase	3
p.Phe909Cys	nonsynonymous	Mtase	2
p.Ala910Val	nonsynonymous	Mtase	3

*Mutations in red were also detected as *de novo* events in overgrowth cases.

Data is derived from COSMIC database (www.sanger.ac.uk/cancergenome/COSMIC). Only confirmed somatic mutations in haematological neoplasms (excluding synonymous changes) are included.

Abbreviations: PWWP, proline-tryptophan-tryptophan-proline; ADD, ATRX, DNMT3A, DNMT3L-type zinc finger; Mtase, methyltransferase

Supplementary Figure 1. Thirteen *de novo DNMT3A* mutations in overgrowth cases



*indicates that the reverse sequencing trace is presented

Supplementary Note

The Childhood Overgrowth (COG) Collaboration

The following are members of the Childhood Overgrowth Collaboration and undertook recruitment of cases, samples and clinical information.

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