



HAL
open science

Multiple endocrine neoplasia type 1 caused by mosaic mutation: clinical follow-up and genetic counseling?

Lucie Coppin, Sophie Giraud, Eric Pasmant, Arnaud Lagarde, Marie-Odile North, Lauriane Le-Collen, Valérie Aubert, Grégory Mougel, Miriam Ladsous, Alyzée Louboutin, et al.

► **To cite this version:**

Lucie Coppin, Sophie Giraud, Eric Pasmant, Arnaud Lagarde, Marie-Odile North, et al.. Multiple endocrine neoplasia type 1 caused by mosaic mutation: clinical follow-up and genetic counseling?. European Journal of Endocrinology, 2023, 187 (1), pp.K1-K6. 10.1530/EJE-22-0171 . hal-04014019

HAL Id: hal-04014019

<https://amu.hal.science/hal-04014019>

Submitted on 3 Mar 2023

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

Multiple endocrine neoplasia type 1 caused by mosaic mutation: what about clinical follow up and genetic counseling?

Lucie Coppin¹, Sophie Giraud², Eric Pasmant^{3,4}, Arnaud Lagarde⁵, Marie-Odile North³,
Lauriane Le-Collen^{6,7}, Valérie Aubert⁸, Grégory Mougél², Miriam Ladsous⁹, Alyzée
Louboutin¹⁰, Hedia Brix¹¹, Magalie Haissaguerre¹², Nicolas Schreyer¹³, Marc Klein¹³, Antoine
Tabarin¹², Brigitte Delemer⁶, Anne Barlier⁵, Marie-Françoise Odou^{14,15}, Pauline Romanet⁵.

Affiliations

¹ Univ. Lille, CNRS, Inserm, CHU Lille, UMR9020-U1277 - CANTHER - Cancer - Heterogeneity Plasticity and Resistance to Therapies, F-59000, Lille, France; Lucie.verstraete@chru-lille.fr

² Genetics Department, Hospices Civils de LYON (HCL), University Hospital, East Pathology Center, LYON. sophie.giraud@chu-lyon.fr

³ Service de Génétique et Biologie Moléculaires, Hôpital Cochin, DMU BioPhyGen, Assistance Publique-Hôpitaux de Paris, AP-HP. Centre-Université de Paris, Paris, France. eric.pasmant@aphp.fr, marie-odile.north@aphp.fr

⁴ Institut Cochin, Inserm U1016, CNRS UMR8104, Université de Paris, CARPEM, Paris, France.

⁵ Aix Marseille Univ, APHM, INSERM, MMG, Laboratory of Molecular Biology Hospital La Conception, Marseille, France; arnaud.lagarde@ap-hm.fr, gregory.mougel@ap-hm.fr, anne.barlier@univ-amu.fr, pauline.romanet@univ-amu.fr

⁶ Endocrinology, Diabetology and Nutrition Unit, University Hospital of Reims, Reims, France; l-le-collen@chu-reims.fr, bdelemer@chu-reims.fr

⁷ Inserm/CNRS UMR 1283/8199, Pasteur Institute of Lille, EGID, Lille, France

⁸ Cabinet médical, Rue de Sarre, METZ dr.valerie.aubert@orange.fr

⁹ CHU Lille, Service d'Endocrinologie, Diabétologie, Métabolisme et Nutrition, Hôpital Claude Huriez, F-59000 Lille, France miriam.ladsous@chu-lille.fr

¹⁰ Hôpital de Quimper alizee.louboutin@yahoo.fr

¹¹ Department of Gastroenterology and Digestive Oncology, Reims University Hospital, Reims, France; hbrix@chu-reims.fr

¹² Service d'Endocrinologie, Centre Hospitalier Universitaire, Hôpital du Haut Levêque, Pessac, France; magalie.haissaguerre@chu-bordeaux.fr, antoine.tabarin@chu-bordeaux.fr

35 ¹³Service endocrinologie, CHU de Nancy, hôpital de Brabois, Vandoeuvre-lès-Nancy, France;
36 m.klein@chu-nancy.fr, N.SCHEYER@chru-nancy.fr

37 ¹⁴CHU Lille, Service de Biochimie et Biologie moléculaire « Hormonologie, Métabolisme-
38 Nutrition, Oncologie », Lille, France ; mf.paris@chu-lille.fr

39 ¹⁵Univ. Lille, Inserm, CHU Lille, U1286 – Infinite – Institute for Translational Research in
40 Inflammation, F-59000 Lille, France

41

42 **Correspondence:**

43 Dr Pauline ROMANET, MD, PhD

44 Aix Marseille Univ, INSERM, MMG, UMR 1251

45 Faculté des sciences médicales et paramédicales,

46 27, boulevard Jean Moulin 13385 Marseille cedex 5, France.

47 Tel +33 491 69 87 89

48 Fax +33 491 69 89 20

49 pauline.romanet@univ-amu.fr

50

51 **Word Count** : 1619

52 **Running title:** MEN1 caused by mosaic mutation

53 **Key words:** MEN1, mosaicism, NGS, next generation sequencing, mosaic, genetic testing,

54 hyperparathyroidism, thymic NET

55

56

57

58

59

60 **Abstract (136/250):**

61 MEN1 is an autosomal dominant hereditary syndrome characterized by several endocrine
62 tumors, in most cases affecting the parathyroid glands, pancreas, and anterior pituitary. It is
63 the result of inactivating mutations in the tumor suppressor gene *MEN1*. More than 1300
64 different mutations have been identified in this gene. Mosaic *MEN1* mutations have been
65 previously described in only a few patients in the literature. In this paper we provide a review
66 of six cases of *MEN1* mosaicism reported in the literature supplemented with six additional
67 cases described by the French TENgen network of laboratories. This review highlights that (i)
68 *MEN1* mosaicism is not associated with a mild phenotype, (ii) results in the same natural
69 history as heterozygous *MEN1* mutation, and (iii) that more systematic detection of *MEN1*
70 mosaic mutation enables improvements in both patient monitoring and genetic counseling.

71

72

73

MAIN TEXT

74 **BACKGROUND**

75 Multiple endocrine neoplasia type 1 (MEN1) is a rare disease caused by inactivating
76 mutations in the *MEN1* gene (1). MEN1 is characterized by a broad spectrum of clinical
77 manifestations from which the three cardinal lesions are primary hyperparathyroidism (PHPT),
78 pituitary neuroendocrine tumors (PitNETs), and neuroendocrine duodeno-pancreatic
79 tumors (2). Other neuro-endocrine tumors (NETs) such as adrenal cortical tumors,
80 bronchopulmonary NETs, or thymic NETs may also be associated. Twenty-eight to 70% of

81 MEN1 patients die due to the disease, particularly due to the pancreatic and carcinoid lesions
82 (3). In 2012 a group of experts provided guidelines for the evaluation, treatment, and genetic
83 testing of MEN1 disease (4). MEN1 is diagnosed based on clinical criteria in all patients with
84 two or more MEN1-related tumors, on family criteria in patients having one MEN1-related
85 tumor and a first-degree relative with MEN1, and on genetic criteria in all patients,
86 symptomatic or not, who carry a *MEN1* pathogenic variant. The MEN1 diagnosis allows
87 patients to benefit from a multidisciplinary follow-up program which enables early detection
88 and treatment of lesions (4, 5).

89 *MEN1* is a tumor suppressor gene (1, 6) and *MEN1* pathogenic or likely pathogenic
90 variants are detected in 90% to 95% of family cases, and in 30-45% of sporadic cases bearing
91 the classical triad (7, 8, 9). A false-negative genetic test can occur in cases where a *de novo*
92 mutation occurs during postzygotic development, that is, after fertilization. In these cases,
93 only a proportion of cells in the organism harbor the variation; a feature termed mosaicism or
94 somatic mosaicism. In mosaicism, germline genetic testing may be negative if the fraction of
95 mutated alleles in the blood is below the detection threshold. Only a few cases of *MEN1*
96 mosaicism have been reported in the literature (10, 11, 12, 13, 14). The impact of *MEN1*
97 mosaicism on patient phenotype and on the natural history of the disease is unknown. Here
98 we review all published cases of mosaicism and additionally consider six unpublished cases
99 from the TENgen group (French network of laboratories working on NETs), in order to clarify
100 the clinical presentation and natural history of *MEN1* mosaicism and then discuss appropriate
101 genetic counseling.

102

103 REPORT

104 To date, only 6 cases of *MEN1* mosaicism have been reported in the literature (see
105 Table). The first description was published by Klein *et al* in 2005 (10) who used molecular
106 subcloning to identify a *MEN1* truncating mutation with a 10 % allele frequency in a 27 year-
107 old woman who presented all typical *MEN1* lesions (PHPT, prolactinoma, and a pancreatic
108 lesion, patient #1). The second case (patient #2), a woman diagnosed with PHPT at age 52,
109 was reported by Farook *et al* in 2011(11). She presented several recurrences of PHPT and her
110 daughter also developed PHPT, leading to genetic testing. The index case later developed a
111 pituitary microadenoma and two pancreatic lesions. A mosaic mutation in *MEN1* was
112 identified at a level of 10% and the same mutation found in her daughter at the heterozygous
113 level. Mauchlen *et al* also reported a case of *MEN1* mosaicism with 15 % of mutated alleles in
114 a 43-year-old patient with a parathyroid adenoma and pancreatic gastrinoma (patient #3)
115 (12).

116 The three other cases were reported in relatively older patients. Two were identified
117 by Coppin *et al* (13) using NGS. The first was a patient who developed PHPT at age 57 and a
118 pancreatic NET at age 66 (patient #4). The mutation (6 % mutated alleles) had previously been
119 identified at the heterozygous level in her daughter. The second case was a 68-year-old
120 patient with hyperparathyroidism and a PitNET (patient #5) with 15 % mutated alleles. Later
121 Beijers *et al* described an entire *MEN1* gene mosaic deletion, using MLPA, in a 63-year-old
122 patient with PHPT and metastatic pancreatic NETs (patient #6, 14). Again, the deletion had
123 previously been identified at a heterozygous level in his 2 children.

124 In addition to cases from the literature review, we present here 6 new cases of *MEN1*
125 mosaicism identified by next-generation sequencing (NGS) during recent years in the French
126 TENgen network. Three were identified in young patients. The first showed a severe clinical
127 presentation: the patient developed PHPT at age 28, an endocrine thymic tumor at age 35 and

128 a pituitary adenoma at age 37 (patient #7). The proportion of mutated alleles was 6%. The
129 second case was a 24-year-old man harboring a *MEN1* mosaic mutation at a level of 10 %
130 (patient #8). He presented with PHPT and a duodenopancreatic NET and developed a PitNET
131 at age 27. In the third case, a *MEN1* mosaic mutation was found in a 24-year-old man at a level
132 of 11 % with a milder clinical presentation, only a PitNET being diagnosed (patient #9).

133 Another *MEN1* mosaic mutation was identified in an older woman first reported in
134 2013 by Cuny et al (15) who developed PHPT and a PitNET before age 45. Her daughter was
135 diagnosed with a heterozygous frameshift insertion of 26 nucleotides in *MEN1*, discovered
136 after she developed a macroprolactinoma at age 13 and PHPT at age 20. Using NGS we
137 retrospectively identified the same truncating mutation as in her child, at a level of 6.3 %
138 mutated alleles (patient #10).

139 We also detected *MEN1* mosaicism in two older patients with severe MEN1. The first
140 patient presented with PHPT, a PitNET and duodenopancreatic NETs at age 46 and an
141 endocrine thymic tumor was diagnosed a year later (patient #11). The proportion of mutated
142 alleles identified by NGS was 2.5%. Additionally, *MEN1* mosaicism was identified at 9.4% in a
143 patient who presented at age 43 with PHPT and a metastatic thymic NET. He later developed
144 duodenopancreatic NETs at age 53 (patient #12).

145 We compared the age of occurrence of the first, second and third lesions of the classic
146 MEN1 triad (PHPT , PitNET, and duodeno-pancreatic NET) between these 12 patients with
147 *MEN1* mosaicism and the 497 index cases with pathogenic and likely pathogenic variants from
148 the UMD-MEN1 database as previously described (16). No difference in the cumulative
149 incidence of lesions was found (Figure 1).

150

151

152

153 DISCUSSION

154 Only six cases of *MEN1* mosaicism have been reported in the literature to date (10, 11,
155 12, 13, 14). We report here six new patients, thus constituting the largest series ever reported.
156 Some studies have suggested that the *MEN1* phenotype may be attenuated in patients with
157 *MEN1* mosaicism (11). This review clearly demonstrates that patients with *MEN1* mosaicism
158 can develop the classical triad (patients #1, #3, #4), even early in life (patients #1, #4). Patients
159 with *MEN1* mosaicism appear to develop the classic *MEN1* lesions at the same age as others
160 (Figure 1). Moreover *MEN1* can become a very aggressive disease with multiple lesions (6
161 patients reported here showed at least 3 *MEN1* lesions) or show metastatic lesions (patients
162 #6, #12) even in patients harboring *MEN1* mosaicism. For this reason careful monitoring in
163 these patients must not be relaxed.

164 In terms of the nature of the variant, we observed that all types of alterations were
165 represented in patients with mosaicism (5/12 frameshift variants, 3/12 non-sense variants, 2
166 variants affecting splice junctions, 1 entire gene deletion, 1 missense variant). A relationship
167 between mutated allele frequency and the severity of the disease was not established. In
168 patient #11, a very low mutated allele frequency of 2.5% was responsible for disease affecting
169 multiple tissues, whereas mosaicism at 10% can cause complete *MEN1* disease at a young age.
170 For the moment, there is no argument in favor of modifying the follow-up program of *MEN1*
171 patients with mosaicism in relation to either age of onset or frequency.

172 *MEN1* mosaicism can be discovered in index cases with a symptomatic child harboring
173 a heterozygous mutation (patient #2, #4, #6, and #10). First-line negative genetic screening by
174 Sanger sequencing could lead to the conclusion of a phenocopy in the symptomatic parent
175 and the exclusion of other siblings from presymptomatic genetic screening. We suggest that

176 physicians should consider that a symptomatic parent with a child presenting with an
177 apparently *de novo* mutation may harbor a mosaic mutation until proven otherwise. We note
178 that in the literature there is only one symptomatic parent of a child carrying a *MEN1*
179 mutation, in whom genetic explorations were negative, even by deep NGS (17). This man
180 presented with hypercalcemia and elevated PTH levels (10.9 mg/mL, 80 pg/mL (normal assay
181 range 14–72)) at age 63, but imaging data (ultrasound and Sestamibi parathyroid scan)
182 provided no information. The analysis of abnormal tissue in this case could have made it
183 possible to definitively exclude mosaicism.

184 *MEN1* mosaicism can also be identified in sporadic index cases bearing one or several
185 *MEN1* lesions. Success in identifying *MEN1* mosaicism in these patients is really fortunate for
186 the patient and his/her family. Firstly, it allows confirmation of the *MEN1* diagnosis in the
187 patient, even in a patient with an isolated PitNET (patient #9), or late multiple lesions (patient
188 #5). Additionally it clarifies genetic counseling. Since the disease is a mosaic, it confirms the
189 appearance of the disease in the patient. Consequently, the patient's parents and siblings can
190 be reassured and excluded from a *MEN1* follow-up program. Presymptomatic genetic
191 screening can also be started for the patient's children. In fact, the component of genetic
192 counseling that addresses the risk of recurrence remains challenging in mosaic genetic
193 diseases, since it depends on the proportion of germ cell progenitors that harbor the
194 mutation. One way in which to determine this would be to genotype the sperm of male
195 patients (18, 19). However, no study has thus far been conducted to determine the reliability
196 of a negative result. Nevertheless, as NGS can detect mosaic variants at low AF (13, 20, 21,
197 22, 23); we suggest geneticists and physicians keep in mind that mosaicism is likely
198 underestimated in unresolved *MEN1* patients.

199

200

201 **Author Information:** The authors declare no conflict of interest.

202

203 **Funding:** All phases of this study were supported by grants from the Institut National de lutte
204 contre le Cancer (INCa), the MarMaRa Institute, and the French Ministry of Health. The project
205 resulting in this publication received funding from the Excellence Initiative of Aix Marseille
206 University -A*Midex- a French "Investissement d'Avenir" - Institute MarMaRa AMX-19-IET-
207 007.

208

209 **Ethics declaration:** Informed consent was obtained from the subjects involved in the study.

210 The study was conducted according to the guidelines of the Declaration of Helsinki and the UE
211 General Data Protection Regulation.

212

213 **REFERENCES:**

214 1. Chandrasekharappa SC, Guru SC, Manickam P, Olufemi SE, Collins FS, Emmert-Buck MR,
215 Debelenko L V, Zhuang Z, Lubensky IA, Liotta LA, Crabtree JS, Wang Y, Roe BA,
216 Weisemann J, Boguski MS, Agarwal SK, Kester MB, Kim YS, Heppner C, Dong Q, Spiegel
217 AM, Burns AL, & Marx SJ. Positional cloning of the gene for multiple endocrine
218 neoplasia-type 1. *Science (New York, N.Y.)* 1997 **276** 404–407.

219 2. Wermer P. Genetic aspects of adenomatosis of endocrine glands. *The American Journal*
220 *of Medicine* 1954 **16** 363–371. (doi:10.1016/0002-9343(54)90353-8)

221 3. Goudet P, Murat A, Biquet C, Cardot-Bauters C, Costa A, Ruzniewski P, Niccoli P,
222 Ménégaux F, Chabrier G, Borson-Chazot F, Tabarin A, Bouchard P, Delemer B, Beckers

- 223 A, & Bonithon-Kopp C. Risk Factors and Causes of Death in MEN1 Disease. A GTE
224 (Groupe d'Etude des Tumeurs Endocrines) Cohort Study Among 758 Patients. *World*
225 *Journal of Surgery* 2010 **34** 249–255. (doi:10.1007/s00268-009-0290-1)
- 226 4. Thakker R V, Newey PJ, Walls G V, Bilezikian J, Dralle H, Ebeling PR, Melmed S, Sakurai
227 A, Tonelli F, & Brandi ML. Clinical practice guidelines for multiple endocrine neoplasia
228 type 1 (MEN1). *The Journal of clinical endocrinology and metabolism* 2012 **97** 2990–
229 3011. (doi:10.1210/jc.2012-1230)
- 230 5. Lourenço-Jr DM, Toledo RA, Coutinho FL, Margarido LC, Siqueira SAC, Santos MACG d,
231 Montenegro FL de M, Machado MCC, Toledo SPA, Lourenço DM, Toledo RA, Coutinho
232 FL, Margarido LC, Siqueira SAC, Santos MACG dos, Montenegro FL de M, Machado MCC,
233 & Toledo SPA. The impact of clinical and genetic screenings on the management of the
234 multiple endocrine neoplasia type 1. *Clinics* 2007 **62** 465–470. (doi:10.1590/S1807-
235 59322007000400014)
- 236 6. Marx S, Spiegel AM, Skarulis MC, Doppman JL, Collins FS, & Liotta LA. Multiple
237 Endocrine Neoplasia Type 1: Clinical and Genetic Topics. *Annals of Internal Medicine*
238 1998 **129** 484. (doi:10.7326/0003-4819-129-6-199809150-00011)
- 239 7. Lemos MC & Thakker R V. Multiple endocrine neoplasia type 1 (MEN1): analysis of 1336
240 mutations reported in the first decade following identification of the gene. *Human*
241 *mutation* 2008 **29** 22–32. (doi:10.1002/humu.20605)
- 242 8. Odou MF, Cardot-Bauters C, Vantyghem MC, Carnaille B, Leteurtre E, Pigny P, Verrier-
243 Mine O, Desaillood R, & Porchet N. Contribution of genetic analysis in screening for
244 MEN1 among patients with sporadic disease and one or more typical manifestation.

- 245 *Annales d'endocrinologie* 2006 **67** 581–587.
- 246 9. Tham E, Grandell U, Lindgren E, Toss G, Skogseid B, & Nordenskjöld M. Clinical testing
247 for mutations in the MEN1 gene in Sweden: a report on 200 unrelated cases. *The*
248 *Journal of clinical endocrinology and metabolism* 2007 **92** 3389–3395.
249 (doi:10.1210/jc.2007-0476)
- 250 10. Klein RD, Salih S, Bessoni J, & Bale AE. Clinical testing for multiple endocrine neoplasia
251 type 1 in a DNA diagnostic laboratory. *Genetics in medicine : official journal of the*
252 *American College of Medical Genetics* 2005 **7** 131–138.
253 (doi:10.109701.GIM.0000153663.62300.F8)
- 254 11. Farook S, Kannappan D, Kenz S, Lalloo F, Trainer P, & Brabant G. MEN-1 mosaic: the
255 founder of a family. *Endocrine Abstracts* 2011 **25** .
- 256 12. Mauchlen R, Carty D, Talla M, & Drummond R. Multiple endocrine neoplasia type 1
257 (MEN1) mosaicism caused by a c.124G>A variant in the MEN1 gene. *Endocrine*
258 *Abstracts* 2019 **65** . (doi:10.1530/ENDOABS.65.CC4)
- 259 13. Coppin L, Ferrière A, Crépin M, Haissaguerre M, Ladsous M, Tabarin A, & Odou MF.
260 Diagnosis of mosaic mutations in the MEN1 gene by next generation sequencing.
261 *European journal of endocrinology* 2019 **180** L1–L3. (doi:10.1530/EJE-18-0852)
- 262 14. Beijers HJBH, Stikkelbroeck NML, Mensenkamp AR, Pfundt R, Luijt RB Van Der, Luijt RB
263 van der, Timmers HJLM, Hermus ARMM, & Kempers MJE. Germline and somatic
264 mosaicism in a family with multiple endocrine neoplasia type 1 (MEN1) syndrome.
265 *European journal of endocrinology* 2019 **180** 1–5. (doi:10.1530/EJE-18-0778)

- 266 15. Cuny T, Pertuit M, Sahnoun-Fathallah M, Daly A, Occhi G, Odou MF, Tabarin A, Nunes
267 ML, Delemer B, Rohmer V, Desailoud R, Kerlan V, Chabre O, Sadoul JL, Cogne M, Caron
268 P, Cortet-Rudelli C, Lienhardt A, Raingeard I, Guedj AM, Brue T, Beckers A, Weryha G,
269 Enjalbert A, & Barlier A. Genetic analysis in young patients with sporadic pituitary
270 macroadenomas: besides AIP don't forget MEN1 genetic analysis. *European journal of*
271 *endocrinology* 2013 **168** 533–541. (doi:10.1530/EJE-12-0763)
- 272 16. Romanet P, Mohamed A, Giraud S, Odou MF, North MO, Pertuit M, Pasmant E, Coppin
273 L, Guien C, Calender A, Borson-Chazot F, Bérout C, Goudet P, & Barlier A. UMD-MEN1
274 Database: An Overview of the 370 *MEN1* Variants Present in 1676 Patients From the
275 French Population. *The Journal of Clinical Endocrinology & Metabolism* 2019 **104** 753–
276 764. (doi:10.1210/jc.2018-01170)
- 277 17. Laitman Y, Jaffe A, Schayek H, & Friedman E. De novo mutation in MEN1 is not
278 associated with parental somatic mosaicism. *Endocrine-related cancer* 2017 **24** L1–L3.
279 (doi:10.1530/ERC-16-0446)
- 280 18. Pasmant E & Pacot L. Should we genotype the sperm of fathers from patients with 'de
281 novo' mutations? *European Journal of Endocrinology* 2020 **182** C1–C3.
282 (doi:10.1530/EJE-19-0759)
- 283 19. Lin Y, Cai Y, Xu J, Zeng C, Sheng H, Yu Y, Li X, & Liu L. 'Isolated' germline mosaicism in
284 the phenotypically normal father of a girl with X-linked hypophosphatemic rickets.
285 *European Journal of Endocrinology* 2020 **182** K1–K6. (doi:10.1530/EJE-19-0472)
- 286 20. Coppin L, Plouvier P, Crépin M, Jourdain AS, Ait Yahya E, Richard S, Bressac-de Paillerets
287 B, Cardot-Bauters C, Lejeune S, Leclerc J, & Pigny P. Optimization of Next-Generation

- 288 Sequencing Technologies for von Hippel Lindau (VHL) Mosaic Mutation Detection and
289 Development of Confirmation Methods. *The Journal of molecular diagnostics : JMD*
290 2019 **21** 462–470. (doi:10.1016/j.jmoldx.2019.01.005)
- 291 21. Coppin L, Grutzmacher C, Crépin M, Destailleur E, Giraud S, Cardot-Bauters C, Porchet
292 N, & Pigny P. VHL mosaicism can be detected by clinical next-generation sequencing
293 and is not restricted to patients with a mild phenotype. *European Journal of Human*
294 *Genetics* 2014 **22** 1149–1152. (doi:10.1038/ejhg.2013.279)
- 295 22. Izawa K, Hijikata A, Tanaka N, Kawai T, Saito MK, Goldbach-Mansky R, Aksentijevich I,
296 Yasumi T, Nakahata T, Heike T, Nishikomori R, & Ohara O. Detection of Base
297 Substitution-Type Somatic Mosaicism of the NLRP3 Gene with >99.9% Statistical
298 Confidence by Massively Parallel Sequencing. *DNA Research* 2012 **19** 143–152.
299 (doi:10.1093/dnares/dsr047)
- 300 23. Evans DG, Hartley CL, Smith PT, King AT, Bowers NL, Tobi S, Wallace AJ, Perry M, Anup
301 R, Lloyd SKW, Rutherford SA, Hammerbeck-Ward C, Pathmanaban ON, Stapleton E,
302 Freeman SR, Kellett M, Halliday D, Parry A, Gair JJ, Axon P, Laitt R, Thomas O, Afridi SK,
303 Obholzer R, English Specialist NF research group D, Duff C, Stivaros SM, Vassallo G,
304 Harkness EF, & Smith MJ. Incidence of mosaicism in 1055 de novo NF2 cases: much
305 higher than previous estimates with high utility of next-generation sequencing.
306 *Genetics in medicine : official journal of the American College of Medical Genetics* 2020
307 **22** 53–59. (doi:10.1038/s41436-019-0598-7)
- 308
309

310

311

312 Figure 1: Cumulative incidence of the first, second and third MEN1 cardinal lesions in patients
313 with MEN1 mosaicism and in index cases from the UMD-MEN1 database.

314 A total of 497 index cases with heterozygous MEN1 (likely) pathogenic variants, and for which
315 the age of occurrence of the lesions of the classic MEN1 triad was known were included.

316 Patients from the UMD MEN1 database with incomplete clinical data were excluded. A) Age
317 at occurrence of the first MEN1 cardinal lesion. B) Age at occurrence of the second MEN1
318 cardinal lesion. C) Age at occurrence of the third MEN1 cardinal lesion.

319 Log rank test performed with Prism V9 (Graphpad) showed no difference between the 2
320 groups for the 3 conditions.

321

322

323

324

325