The progressive fragmentation of the KIT/PDGFRA wild-type (WT) gastrointestinal stromal tumors (GIST)

Margherita Nannini1*, Milena Urbini2, Annalisa Astolfi2, Guido Biasco1,2 and Mara A. Pantaleo1,2

Abstract

Recent advances in molecular biology have revolutionized the concept of KIT/PDGFRA wild type (WT) gastrointestinal stromal tumors (GIST) than the past. Indeed, from being defined as GIST without KIT or PDGFRA mutations, we are now faced with the opposite scenario, where KIT/PDGFRA WT GIST are “positively” defined according to their specific molecular alterations. In particular, if until recently KIT/PDGFRA GIST without abnormalities of KIT, PDGFRA, SDH, and the RAS signaling pathway were referred as quadruple WT GIST, today also this small subset of GIST is emerging out as a group of heterogeneous distinct entities with multiple different molecular alterations. Therefore, given this still growing and rapidly evolving scenario, the progressive molecular fragmentation may inevitably lead over the time to the disappearance of KIT/PDGFRA WT GIST, destined to be singularly defined by their molecular fingerprint.

Keywords: GIST, SDH, Quadruple wild-type, ETV6–NTRK3, MEN1, MAX, FGFR1

Gastrointestinal stromal tumors (GIST) that lack KIT or platelet-derived growth factor receptor alpha (PDGFRA) mutations, that are around 10–15% of all cases, have always been classified as KIT/PDGFRA wild type GIST, short-named WT GIST [1]. Only recently, a more comprehensive molecular analysis have shown that KIT/PDGFRA WT GIST are a rather heterogeneous group of different diseases than one single entity [2].

20–40% of all KIT/PDGFRA WT GIST are succinate dehydrogenase complex (SDH)-deficient GIST, recognized by the loss of subunit B (SDHB) protein expression most often due to germ-line and/or somatic loss-of-function mutations in any of the four SDH subunits (A, B, C, or D). The SDH-deficient GIST share a pathognomonic profile characterized by young age, female gender predilection, gastric localization, mixed epithelioid and spindle cell morphology, diffuse KIT and ANO1 (DOG1) IHC positivity, frequent lymph node metastatic involvement, and an indolent behaviour even often metastatic up-front [3–9]. Moreover, SDH-deficient GIST are characterized by the over-expression of the insulin growth factor 1 receptor (IGF1R) [10, 11]. Finally, they also display a common epigenomic background, distinguished by a distinctive hypermethylation and miRNA profile [12–17]. In particular, SDH-deficient GIST present a marked hypermethylation profile, generally implicates the Krebs cycle as SDH-mutant paraganglioma and pheochromocytoma (Pgl/Pheo) [12]. Moreover, SDH-deficient GIST also show a distinctive miRNA expression profile characterized by miR-139-5p, 455-5p and let-7b signature, that may represent the epigenetic modulator of IGF1R expression and then a potential onco-miR mark of this subset of GIST [17].

The subgroup of the remaining KIT/PDGFRA WT GIST, but not SDH-deficient, have been further characterized: 4–13% carry a BRAF V600E mutation, are localized more frequently in small intestine and seem to have a more favourable prognosis [18–21]. Within the not SDH-deficient, some GIST have a neurofibromatosis (NF) type 1 mutation and show a female prevalence, a frequent non-gastric site and multifocal localization often unveiling an unrecognized NF1 syndromic condition [22–26].
Half of the KIT or PDGFRA WT GIST have been identified to be either SDH-deficient or BRAF/RAS/NF1 mutated, but the other half has still remains orphan of a specific molecular event and thus has been named as quadruple WT-GIST [27]. However, the transcriptome profile of this small subgroup is so profoundly different from the other GIST, either KIT/PDGFRA WT or -mutated GIST, that quadruple WT GIST could represent another unique group within the family of GIST [28].

Nevertheless, recently it has been shown that quadruple WT GIST have a greater molecular heterogeneity, with many different and probably mutually exclusive mutational events (Fig. 1). The presence of an ETV6-NTRK3 gene fusion has been firstly described in a case of rectal quadruple WT GIST [29]. The same translocation have been also reported in a colon primary quadruple WT GIST [30]. Moreover, two fusion genes involving FGFR1 were reported in three cases of quadruple WT GIST (FGFR1–HOOK3 and FGFR1–TACC1) [30]. Recently, other fusion events (KIT–PDGFRA, MARK2-PFIA1 and SPRED2-NELFCD) have also been detected [31, 32]. Finally, relevant somatic mutations, including TP53, MEN1, MAX, CHD4, FGFR1, CTDNN2, CBL, ARID1A, BCOR and APC were also identified [30–33]. Interestingly, MEN1 and MAX mutations, along with NF1 and SDH, further extend the list of genes detected in KIT/PDGFRA WT GIST, genes which are characteristic for neuroendocrine tumors. Moreover, detection of high expression level of genes involved in the neural commitment process, such ASCL1 and EPHA4, further support the hypothesis of a neuroendocrine like signature for some quadruple WT GIST [32].

Given the recent findings, also quadruple WT GIST, the small subset of GIST that lack abnormalities of KIT, PDGFRA, SDH, and the RAS signalling pathway, can be considered as a group of heterogeneous single entities with different molecular alterations. Therefore, a different scenario than expected is emerging. Despite their unquestionable GIST morphology, given this marked molecular heterogeneity, quadruple WT GIST could be a different disease than GIST. Otherwise, trusting unquestionable GIST morphology, it could be argued that quadruple WT GIST may arise from a distinct population of pluripotent interstitial cells of Cajal (ICC) [34, 35], or that they may share a molecular driver at the epigenomic level, given their homogeneous gene expression profile.

If in the past KIT/PDGFRA WT GIST has been for long “negatively” defined by exclusion, we are now faced with the opposite scenario, where KIT/PDGFRA WT GIST are “positively” defined according to their specific molecular alterations. Over the time, this is inevitably leading to a progressive fragmentation of the KIT/PDGFRA WT GIST, until make them disappearing.
Abbreviations
GIST: gastrointestinal stromal tumors; PDGFRA: platelet-derived growth factor receptor alpha; WT: wild type; NF1: neurofibromatosis type 1; ICC: interstitial cell of Cajal; SDH: succinate dehydrogenase.

Authors’ contributions
MN has made substantial contributions to the conception of the manuscript and drafted the final manuscript. MU has carried out the figure. AA, GB, MAP have been involved in revising the manuscript critically for important intellectual content and have given final approval of the version to be published. All authors read and approved the final manuscript.

Author details
1 Department of Specialized, Experimental and Diagnostics Medicine, Sant’Orsola-Malpighi Hospital, University of Bologna, Via Massarenti 9, Bologna, Italy. 2 “Giorgio Pordi” Cancer Research Center, University of Bologna, Bologna, Italy.

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References
1. Corless CL, Fletcher JA, Heinrich MC. Biology of gastrointestinal stromal tumors. J Clin Oncol. 2004;22:3813–25.
2. Nannini M, Bisaco G, Astolfi A, Pantaleo MA. An overview on molecular biology of KIT/PDGFRA wild-type (WT) gastrointestinal stromal tumors (GIST). J Med Genet. 2013;50:653–61.
3. Gill AJ, Chou A, Vilain R, Clarkson A, Lui M, Jin R, Tobias V, Samra J, Goldstein D, Smith C, Sioson L, Parker N, Smith RC, Sywak M, Sidhu SB, Wyatt JM, Robinson BG, Eckstein RP, Benn DE, Clifton-Bagh RJ. Immunohistochemistry of SDHB divides gastrointestinal stromal tumors (GISTs) into 2 distinct types. Am J Surg Pathol. 2010;34:636–44.
4. Janeway KA, Kim SY, Lodish M, Novey V, Rustin P, Jaffe EA, Dhayal L, Liegl B, Ball ER, Raygada M, MaL, Ahl K, Heimrich J, NIH Pediatric and Wild-Type GIST Clinic, O’Sullivan M, de Krigger RR, Dirinjen WN, Demetri GD, Antonescu CR, Fletcher JA, Helman L, Stratakis CA. Defects in succinate dehydrogenase in gastrointestinal stromal tumors lacking KIT and PDGF-R. J Clin Oncol. 2013;31:656–63.
5. Miettinen M, Wang ZF, Sarlomo-Rikala M, Ou S, Rutkowski P, Lasota J. Succinate dehydrogenase-deficient GISTs: a clinicopathological, immunohistochemical, and molecular genetic study of 66 gastric GISTs with predilection to young age. Am J Surg Pathol. 2011;35:1712–21.
6. Pantaleo MA, Astolfi A, Urbini M, Nannini M, Paterini P, Indio V, Saponara M, Formica S, Ceccarelli C, Casadò R, Gatto L, Bertolini F, Santini D, Prini MG, Fiorentino M, Bisacca G, GIST Study Group. Analysis of all subunits, SDHA, SDHB, SDHC, SDHD, of the succinate dehydrogenase complex in KIT/PDGFRA wild-type GIST. Eur J Hum Genet. 2014;22:32–9.
7. Pantaleo MA, Astolfi A, Indio V, Moore R, Thiessen N, Heinrich MC, Gronchi C, Santini D, Catena F, Formica S, Martelli PL, Casadò R, Pession A, Bisacca G. SDHA loss-of-function mutations in KIT/PDGFRA wild-type gastrointestinal stromal tumors identified by massively parallel sequencing. J Natl Cancer Inst. 2011;103:983–7.
8. Miettinen M, Killian JK, Wang ZF, Lasota J, Lau C, Jones L, Walker P, Riedner M, Zhu Y, Kim SY, Helman L, Meltzer P. Immunohistochemical loss of succinate dehydrogenase subunit A (SDHA) in gastrointestinal stromal tumors (GISTs) signals SDHA germline mutation. Am J Surg Pathol. 2013;37:234–40.
9. Pantaleo MA, Lolli C, Nannini M, Astolfi A, Indio V, Saponara M, Urbini M, La Rovere S, Gill A, Goldstein D, Ceccarelli C, Santini D, Rossi G, Fiorentino M, Di Sciascio V, Fusaroli P, Mandrioli A, Gatto L, Catena F, Basso U, Ercolani G, Pinna AD, Bisacca G. Good survival outcome of metastatic SDH-deficient gastrointestinal stromal tumors harboring SDHA mutations. Genet Med. 2015;17:391–5.
10. Nannini M, Astolfi A, Paterini P, Urbini M, Santini D, Catena F, Indio V, Casa- dio R, Pinna AD, Bisacca G, Pantaleo MA. Expression of IGF-1 receptor in KIT/PDGF receptor-a wild-type gastrointestinal stromal tumors with succinate dehydrogenase complex dysfunction. Future Oncol. 2013;9:121–6.
11. Belinsky MG, Rink L, Flieder DB, Jahnemi MS, Schiffman JD, Godwin AK, Mehren MV. Overexpression of insulin-like growth factor 1 receptor and frequent mutational inactivation of SDHA in wild-type SDHB-negative gastrointestinal stromal tumors. Genes Chromosom Cancer. 2013;52:214–24.
12. Killian JK, Kim SY, Miettinen M, Smith C, Merino M, Tsokos M, Quezada M, Smith WT, Jr. Jakhomis MS, Xekouki P, Szarek E, Walker RL, Lasota J, Raffeld M, Klitzle B, Wang Z, Jones L, Zhu Y, Wang Y (Waterfall JJ, O’Sullivan M), Bilikova M, Pacak K, Stratakis C, Janeway KA, Schiffman JD, Fan BJ, Helman L, Meltzer PS. Succinate dehydrogenase mutation underlies global epigenomic divergence in gastrointestinal stromal tumor. Cancer Discov. 2013;3:645–7.
13. Mason EF, Hornick JL. Succinate dehydrogenase deficiency is associated with decreased S-hydroxymethylcytosine production in gastrointestinal stromal tumors: implications for mechanisms of tumorigenesis. Mod Pathol. 2013;26:1492–7.
14. Killian JK, Miettinen M, Walker RL, Wang Y, Zhu YJ, Waterfall JJ, Noyes N, Retnakumar P, Yang Z, Smith WH Jr, Killian MS, Lue CC, Pineda MW, Walling J, Stevenson H, Smith CH, Wang Z, Lasota J, Kim SY, Boioks SA, Helman LJ, Meltzer PS. Recurrent epimutation of SDHC in gastrointestinal stromal tumors. Sci Transl Med. 2014;6:268.
15. Urbini M, Astolfi A, Indio V, Heinrich MC, Corless CL, Nannini M, Ravegnini G, Bisaco G, Pantaleo MA. SDHC methylation in gastrointestinal stromal tumors (GIST): a case report. BMC Med Genet. 2015;16:87.
16. Kelly L, Bryan K, Kim SY, Janeway KA, Killian JK, Schildhaus HU, Miettinen M, Helman L, Meltzer PS, van de Rijn M, Debiec-Rychter M, O’Sullivan M, NIH Pediatric and Wild-Type GIST Clinic. Post-transcriptional dysregulation by miRNAs is implicated in the pathogenesis of gastrointestinal stromal tumor (GIST). PLoS ONE. 2013;8:e64102.
17. Pantaleo MA, Ravegnini G, Astolfi A, Sirneo V, Nannini M, Saponara M, Urbini M, Gatto L, Indio V, Sammarini G, Santini D, Ferrari M, Negri M, Hrelia P, Bisacca G, Angelini S. Integrating miRNA and gene expression profiling analysis revealed regulatory networks in gastrointestinal stromal tumors. Epigenomics. 2016;8:347–66.
18. Agami A, Terracciano LM, Dimhofer S, Tornillo L, Foerster A, Hartmann A, Bihl MP. V600E BRAF mutations are alternative early molecular events in a subset of KIT/PDGFRα wild-type gastrointestinal stromal tumors. J Clin Pathol. 2009;62:613–6.
19. Daniels M, Lurkin I, Pauli R, Erbstösser E, Hildebrandt U, Hellwig K, Zschille U, Lüders P, Krüger G, Knolle J, Stengel B, Prall F, Hertel K, Lobbecke H, Popp B, Theissig F, Wünsch P, Zwartshof E, Agami A, Schneider-Stock R. Spectrum of KIT/PDGFRA/BRAF mutations and Phosphatidylinositol-3-Kinase pathway gene alterations in gastrointestinal stromal tumors (GIST). Cancer Lett. 2011;312:43–54.
20. Rossi S, Gasparotto D, Miceli R, Toffolato L, Gallina G, Scramale E, Marzotto A, Boscatto E, Messerini L, Bacciari I, Mazzoni G, Capella C, Angioli G, Sonzogni A, Sidoni A, Mariani L, Amore P, Gronchi A, Casali PG, Maestro R, Dei Tos AP. KIT, PDGFRA, and BRAF mutational spectrum impacts on the clinical, phenotypic, and genotypic characteristics of KIT/PDGFRA/BRAF gastrointestinal stromal tumors. J Hematol Oncol. 2011;4:64.
21. Bianchi S, Dei Tos AP, Miettinen M, Fetsch JF, Sobin LH, Lasota J. Gastrointestinal stromal tumors in patients with neurofibromatosis 1: a clinicopathologic and molecular genetic study of 45 cases. Am J Surg Pathol. 2006;30:90–6.
24. Bajor J. Gastrointestinal stromal tumors in patients with type 1 neurofibromatosis. Clin Exp Med J. 2009;3:247–54.
25. Salvi PF, Lorenzon L, Caterino S, Antonelli MS, Balducci G. Gastrointestinal stromal tumors associated with neurofibromatosis type 1: a single centre experience and systematic review of the literature including 252 cases. Int J Surg Oncol. 2013;2013:398570.
26. Gasparotto D, Rossi S, Polano M, Tamborini E, Lorenzetto E, Sbaraglia M, Mondello A, Massani M, Lamon S, Bracci R, Mandolesi A, Frate E, Stanzial F, Agaj J, Mazzoleni G, Plioti S, Gronchi A, Dei Tos AP, Maestro R. Quadruple-negative GIST is a sentinel for unrecognized neurofibromatosis type 1 syndrome. Clin Cancer Res. 2017;23:273–82.
27. Pantaleo MA, Nannini M, Corless CL, Heinrich MC. Quadruple wild-type (WT) GIST: defining the subset of GISTs that lack abnormalities of KIT, PDGFRA, SDH, and the RAS signalling pathway. Cancer Med. 2015;4:101–3.
28. Nannini M, Astolfi A, Urbini M, Indio V, Santini D, Heinrich MC, Corless CL, Ceccarelli C, Saponara M, Mandrioli A, Lollì C, Ercolani G, Brandi G, Biasco G, Pantaleo MA. Integrated genomic study of quadruple-WT GIST (KIT/PDGFRA/SDH/RAS pathway wild-type GIST). BMC Cancer. 2014;14:685.
29. Brenca M, Rossi S, Polano M, Gasparotto D, Zanatta L, Racanelli D, Valori L, Lamon S, Dei Tos AP, Maestro R. Transcriptome sequencing identifies ETV6–NTRK3 as a gene fusion involved in GIST. J Pathol. 2016;238:543–9.
30. Shi E, Chmielicki J, Tang CM, Wang K, Heinrich MC, Kang G, Corless CL, Hong D, Fero KE, Murphy JD, Fanta PT, Ali SM, De Siena M, Burgoyne AM, Movva S, Madlensky Heestrand GM, Trent JC, Kurzrock R, Morosini D, Ross JS, Harisemond O, Sicklick JK. FGFR1 and NTRK3 actionable alterations in “Wild-Type” gastrointestinal stromal tumors. J Transl Med. 2016;14:339.
31. Boikos SA, Pappo AS, Killian JK, LaQuaglia MP, Weldon CB, George S, Trent JC, von Mehren M, Wright JA, Schiffman JD, Raygada M, Pacak K, Meltzer PS, Miettinen MM, Stratakis C, Janeway KA, Helman LJ. Molecular subtypes of KIT/PDGFRα wild-type gastrointestinal stromal tumors: a report from the national institutes of health gastrointestinal stromal tumor clinic. JAMA Oncol. 2016;2:922–8.
32. Pantaleo MA, Urbini M, Indio V, Ravegini G, Nannini M, De Luca M, Tarantino G, Angelini S, Gronchi A, Vincenzi B, Grignani G, Colombò C, Fumagalli E, Gatto L, Saponara M, Ianni M, Paterini P, Santini D, Pirini MG, Ceccarelli C, Altamani A, Grupponi E, Renne SL, Collini P, Stacchiotti S, Brandi G, Calsi Pi, Pinna MG, Astolfi A, Biasco G. Genome-wide analyses identifies MEN1 and MAX mutations and a neuroendocrine-like molecular heterogeneity in quadruple WT GIST. Mol Cancer Res. 2017;15:555–62.
33. Belinsky MG, Rink L, Cai KQ, Capuzzi SJ, Hoang Y, Chien J, Godwin AK, von Mehren M. Somatic loss of function mutations in neurofibromin 1 and MYC associated factor X genes identified by exome-wide sequencing in a wild-type GIST case. BMC Cancer. 2015;15:887.
34. Pantaleo MA, Astolfi A, Nannini M, Ceccarelli C, Formica S, Santini D, Heinrich MC, Corless C, Dei Tos AP, Paterini P, Catena F, Maleddu A, Saponara M, Di Battista M, Biasco G. Differential expression of neural markers in KIT and PDGFRα wild-type gastrointestinal stromal tumours. Histopathology. 2011;59:1071–80.
35. Beadling C, Patterson J, Justussen E, Nelson D, Pantaleo MA, Hornick JL, Chacón M, Corless CL, Heinrich MC. Gene expression of the IGF pathway family distinguishes subsets of gastrointestinal stromal tumors wild type for KIT and PDGFRα. Cancer Med. 2013;2:21–31.