THEMATIC REVIEW

Novel targeted therapeutics for MEN2

Sara Redaelli1, Ivan Plaza-Menacho2 and Luca Mologni1
1School of Medicine and Surgery, University of Milano-Bicocca, Monza, Italy
2Spanish National Cancer Research Center (CNIO), Madrid, Spain
Correspondence should be addressed to L Mologni: luca.mologni@unimib.it

This paper is part of a thematic review section on 25 Years of RET and MEN2. The guest editors for this section were Lois Mulligan and Frank Weber

Abstract

The rearranged during transfection (RET) proto-oncogene was recognized as the multiple endocrine neoplasia type 2 (MEN2) causing gene in 1993. Since then, much effort has been put into a clear understanding of its oncogenic signaling, its biochemical function and ways to block its aberrant activation in MEN2 and related cancers. Several small molecules have been designed, developed or redirected as RET inhibitors for the treatment of MEN2 and sporadic MTC. However, current drugs are mostly active against several other kinases, as they were not originally developed for RET. This limits efficacy and poses safety issues. Therefore, there is still much to do to improve targeted MEN2 treatments. New, more potent and selective molecules, or combinatorial strategies may lead to more effective therapies in the near future. Here, we review the rationale for RET targeting in MEN2, the use of currently available drugs and novel preclinical and clinical RET inhibitor candidates.

MEN2: introduction

Mary (fictitious name) is a beautiful and joyful eighteen-year-old girl who enjoys her life. You do not notice anything wrong with her, if not for her slightly too long and slender fingers and limbs. She has multiple endocrine neoplasia type 2 (MEN2) and a marfanoid appearance. However, in a sense, she is lucky: the cause of her disease had been discovered few years before her birth. Now, she is under treatment with a drug that keeps the disease at bay.

MEN2 is a group of hereditary, autosomal dominant syndromes characterized by the occurrence of various endocrine tumors (Schimke 1984). Three MEN2 clinical subtypes are recognized: MEN2A, MEN2B and familial MTC (FMTC). MEN2A patients develop medullary thyroid carcinoma (MTC), pheochromocytoma and parathyroid hyperplasia or adenoma. In contrast, MEN2B patients do not have parathyroid involvement, but often show a number of additional conditions, such as marfanoid features and mucosal neuromas of the lips and the tongue and gastrointestinal manifestations (Marx 2005, Wells et al. 2013). Moreover, MEN2B is generally more aggressive and occurs earlier in life. FMTC patients only develop MTC; thus, some authors view FMTC as a MEN2A variant without adrenal gland involvement. Clinical management of MEN2 patients includes prophylactic thyroidectomy, periodical biochemical screening for urine catecholamine and epinephrine, as well as serum calcium, calcitonin and parathyroid hormone levels and surgical removal of tumors. The etiology of MEN2 syndrome remained obscure until 1993, when Mulligan and coworkers discovered germline mutations of the RET proto-oncogene in MEN2A patients (Mulligan et al. 1993). RET is a signaling co-receptor for neurotrophic factors (Edery et al. 1997). At that time, it was known to be frequently rearranged in papillary thyroid cancers.
(PTC), but its involvement in MEN2 and MTC was not yet clear (Grieco et al. 1990, Santoro et al. 1990). The paper by Mulligan and colleagues opened a new era for specific treatment of these conditions. MEN2A patients carry cysteine-specific mutations in the extracellular domain of RET, that are involved in conformational stability and kinase activity via intramolecular disulfide bridges. These mutations cause covalent dimerization of RET leading to ligand-independent kinase activation (Plaza-Menacho et al. 2006, Mologni 2011, Goodman et al. 2014). Mutations affecting cysteine 634 account for most cases, with C634W being the most prevalent MEN2A mutation, although other cysteines from the cysteine-rich domain (CRD) have been described (Ito et al. 1997). In contrast, the vast majority of MEN2B patients carry a M918T mutation in the catalytic domain of RET. Methionine 918 is located near the kinase activation loop: mutation to threonine causes the opening of the activation loop and induces faster autophosphorylation kinetics (Plaza-Menacho et al. 2014). FMTC can be caused by both extra- and intracellular domains mutations, including cysteine substitutions at the CRD and kinase domain-activating mutations. Of particular interest from a pharmacological standpoint are FMTC mutations at the gatekeeper valine, V804, which render the kinase refractory to inhibition by some inhibitors (Carlomagno et al. 2004, Mologni et al. 2013).

**RET signaling in MEN2**

Molecular dissection of disease-driving mechanisms is fundamental for the development of precise therapeutics able to target the disease causative process while sparing normal functions on the organism. Perturbation of RET signaling by a series of oncogenic events, i.e. single germline/somatic point mutation, gene rearrangement, overexpression or transcriptional upregulation, is a common hallmark in several human cancers. The understanding of the mechanism of action associated with these oncogenic RET ‘variants’ is crucial for the development of more accurate (i.e., targeted) and successful therapeutic strategies to treat patients with RET-positive cancers.

Despite clear genotype–phenotype correlations observed in the cancer syndrome MEN2, the molecular mechanisms linking the different sets of RET mutations with their specific clinical subtypes are far from understood. Work from the last two and a half decades on the genetics and cell signaling of the RET proto-oncogene has described two main mechanisms of action for specific disease-phenotype-associated RET mutations. First, mutations affecting the extracellular cysteine-rich domain (CRD) of RET, associated with MEN2A and FMTC, lead to covalent dimerization and constitutive activation of the receptor (Takahashi et al. 1999). The recent elucidation of the structural architecture of RET extracellular domain in complex with GDNF-GFRα1 showed a composite ligand-binding site, with RET wrapping around the co-receptor/ligand complex (Goodman et al. 2007). A GFRα1-binding hotspot contacts the RET cadherin-like domains, while the CRD contacts both ligand components and makes membrane-proximal homotypic interactions, leading to receptor homodimerization and activation. These CRD-mediated interactions suggest models both for ligand-induced RET activation and for ligand-independent oncogenic dysregulation by MEN2 cysteine mutations (Goodman et al. 2007). Second, mutations affecting the intracellular domain of RET, usually associated with FMTC and always with the MEN2B phenotype, transform the receptor into a monomeric ligand-independent oncoprotein (Santoro et al. 1995). RET MEN2B variants, in particular RET M918T, show not only an altered catalytic activity but also an altered substrate specificity because they preferentially phosphorylate substrates that, contrary to wild-type RET, are usually preferred by cytoplasmic tyrosine kinases such as FAK (Murakami et al. 1999) and SRC (Kato et al. 2002, Encinas et al. 2004, Plaza-Menacho et al. 2011). The transcription factor STAT3 is one such example (Yuan et al. 2004). In addition, RET-MEN2B mutants seem to lack dependency on activation loop Y905 for both cells transformation and signaling (Iwashita et al. 1996, 1999). One plausible explanation about the molecular basis of the disease spectrum is that a different pattern of receptor autophosphorylation displayed by specific RET mutants connects a different set of phospho-tyrosine-mediated downstream signaling pathways and transcriptional programs with specific clinical phenotypes. Recent biochemical, biophysical and structural evaluation of RET M918T and V804M mutants (both targeting the catalytic domain of RET) revealed increased kinetics of autophosphorylation and a more extended activation loop conformation, giving rise to a better intermolecular substrate compared to RET WT (Plaza-Menacho et al. 2014). These data have important implications due to the perturbation of the temporal assembly and specificity of RET signaling complexes. Another interesting observation is that oncogenic RET seems to be heavily internalized and prolonged treatment with specific tyrosine kinase inhibitors (e.g. sorafenib) induced lysosomal degradation.
(Plaza-Menacho et al. 2007). Interestingly, degradation promoted by tyrosine kinase inhibitor (TKI) was apparently higher for RET C634R (MEN2A) than RET M918T (MEN2B). However, blocking HSP90 activity by 17-AAG induced an equally potent degradation of wild-type RET and MEN2-associated RET mutants (Alfano et al. 2010). Altogether these studies highlight the complexity of RET (oncogenic) signaling, in a way that localization of the receptor in specific subcellular compartments, maturation status and the interaction with intracellular and extracellular components are important elements to understand RET function and oncogenic deregulation.

The rationale of a different temporal pattern of receptor activation with specific clinical phenotypes (Plaza-Menacho et al. 2006) has been supported by several studies. In cell-based experiments, RET M918T (MEN2B) triggers elevated levels of PI3K activity (Murakami et al. 1999) and Jun N-terminal kinase activity JNK (Murakami et al. 2002) compared with RET C634R. In addition, RET M918T switches the specificity of the RET tyrosine kinase toward alternatives substrates that interact with Crk and Nck (Bocciardi et al. 1997). The same applies to the interaction with other SH2- or PTB-containing docking/adaptor proteins. For example, RET M918T (MEN2B) showed both enhanced phosphorylation of Y1062 and association with SHC, compared with RET C634R (MEN2A) mutants, resulting in the higher activation of the RAS/ERK1/2 and the PI3K/AKT signaling pathways (Salvatore et al. 2001). Liu and colleagues showed that RET-MEN2B mutants lack phosphorylation at Y1096, directly leading to a decrease in binding of GRB2 to RET, compared with wild-type RET (Liu et al. 1996). Transactivation of STAT3 by RET C634R (MEN2A) is required for cellular transformation in a process that is independent of non-receptor tyrosine kinases JAKs and SRC (Schurigga et al. 2001). On the contrary, activating point mutations targeting the kinase domain, such as RET Y791F and RET S891A (FMTC) implicate JAKs and SRC kinases in the constitutive activation of STAT3 (Plaza Menacho et al. 2005). In another study, oncogenic RET M918T (MEN2B) was shown to interact with and activated STAT3 more strongly than RET C634R (MEN2A) (Yuan et al. 2004). In the same line, oncogenic RET enhanced in cells the phosphorylation levels of FAK activation loop Y576/577 phosphorylation compared to wild-type RET, and kinase domain mutants (RET M918T-MEN2B and RET V804M, a FMTC-associated drug-resistant mutant) induced a more robust phosphorylation than mutants targeting the extracellular domain (e.g., RET C634R, MEN2A). Interestingly, this pattern was mirrored by levels of SRC Y416 kinase activation promoted by RET, indicating a close relationship between levels of RET, FAK, SRC and STAT3 activation (Plaza-Menacho et al. 2011).

Taken all together, these studies indicate that specific sets of signaling pathways are connected with specific sets of MEN2-associated RET mutations. A better understanding of the molecular mechanisms underlying this cancer syndrome ultimately will be necessary to design new therapeutic strategies to treat this disease. Combinatorial therapies targeting RET and other important (disease-specific) downstream effectors may be a real alternative to mono-therapies based on TKIs targeting (oncogenic) RET catalytic activity (see below).

**Targeted treatments for MEN2/MTC**

**Approved drugs**

The spectacular success of imatinib in chronic myeloid leukemia (CML) patients in early 2000s (Kantarjian et al. 2002) indicated that targeted treatment may change a disease history in rationally selected patients. Belonging to the same protein class as the imatinib target ABL1, RET was the next kid on the block. A list of completed and ongoing clinical trials investigating the efficacy of RET inhibitors in familial thyroid cancer is shown in Table 1.

Vandetanib (Caprelsa, ZD6474, Genzyme Corp), an orally available kinase inhibitor active against VEGFR2 and EGFR, was later found to be also a potent RET inhibitor (Carlomagno et al. 2002, Wedge et al. 2002, Vidal et al. 2005). The efficacy and safety of vandetanib in patients with MTC were first evaluated in two phase II studies (Robinson et al. 2010, Wells et al. 2010). In the trial conducted by Robinson and colleagues, vandetanib administered 100mg/daily which resulted in a partial response (PR) in 16% of the patients and in stable disease (SD) for at least 24 weeks in 53% of the treated cases. Wells and colleagues described the use of 300mg/daily as able to induce a PR in 20% of patients and a SD in 53%, with a median progression-free survival (PFS) of 27.9 months. A large multicenter, randomized placebo-controlled crossover phase III study was conducted in a cohort of 331 patients with hereditary and sporadic forms of MTC (ZETA trial, Wells et al. 2012). The patients were randomly divided 2:1 between vandetanib (300mg/daily, n=231) or placebo (n=100) arms. The predicted median PFS for the vandetanib arm was 30.5 months, significantly higher than the 19.3 months observed for placebo. A PR was observed in 45% patients with a predicted median duration of 22 months, while 42% had SD, yielding a disease control rate of 87%. The analysis showed evidence...
Table 1  Clinical trials investigating RET inhibitors in medullary thyroid cancer patients.

<table>
<thead>
<tr>
<th>Phase</th>
<th>Drug</th>
<th>Dose*</th>
<th>Condition</th>
<th>Outcome*</th>
<th>Reference/ID</th>
</tr>
</thead>
<tbody>
<tr>
<td>II</td>
<td>Vandetanib</td>
<td>100 mg/day</td>
<td>FMTC</td>
<td>16% PR, 53% SD</td>
<td>(39)</td>
</tr>
<tr>
<td>II</td>
<td>Vandetanib</td>
<td>150 mg/m²/day</td>
<td>MEN2B</td>
<td>20% PR, 53% SD</td>
<td>(42)</td>
</tr>
<tr>
<td>III</td>
<td>Vandetanib</td>
<td>300 mg/day</td>
<td>MTC</td>
<td>45% PR, 42% SD</td>
<td>(40)</td>
</tr>
<tr>
<td>I/I</td>
<td>Vandetanib</td>
<td>300 mg/day</td>
<td>RET-mutated thyroid cancer</td>
<td>44% ORR</td>
<td>NCT0131206</td>
</tr>
<tr>
<td>I</td>
<td>Cabozantinib</td>
<td>Dose finding</td>
<td>MTC/DMTC</td>
<td>29% PR, 41% SD</td>
<td>(41)</td>
</tr>
<tr>
<td>III</td>
<td>Cabozantinib</td>
<td>140 mg/day</td>
<td>MTC</td>
<td>28% PR</td>
<td>(46)</td>
</tr>
<tr>
<td>II</td>
<td>Sorafenib</td>
<td>400 mg bid</td>
<td>MTC</td>
<td>6% PR, 88% SD</td>
<td>(51)</td>
</tr>
<tr>
<td>II</td>
<td>Sorafenib</td>
<td>400 mg bid</td>
<td>MTC/DMTC</td>
<td>25% PR, 70% SD</td>
<td>(52)</td>
</tr>
<tr>
<td>II</td>
<td>Lenvatinib</td>
<td>24 mg/day</td>
<td>MTC/DMTC</td>
<td>36% PR</td>
<td>(57)</td>
</tr>
<tr>
<td>II</td>
<td>Sunitinib</td>
<td>37.5 mg/day</td>
<td>MTC/DMTC</td>
<td>50% PR</td>
<td>(61)</td>
</tr>
<tr>
<td>II</td>
<td>Sunitinib</td>
<td>50 mg/day</td>
<td>MTC/DMTC</td>
<td>38% PR, 50% SD</td>
<td>(62)</td>
</tr>
<tr>
<td>II</td>
<td>Motesanib</td>
<td>125 mg/day</td>
<td>MTC</td>
<td>2% PR, 81% SD</td>
<td>(74)</td>
</tr>
<tr>
<td>II</td>
<td>Fostamatinib</td>
<td>200 mg bid</td>
<td>Multi-histology</td>
<td>50% SD</td>
<td>(79)</td>
</tr>
<tr>
<td>I/I</td>
<td>Alectinib</td>
<td>Bid, dose finding</td>
<td>RET-mutated thyroid cancer</td>
<td>n.d.</td>
<td>NCT03157128</td>
</tr>
<tr>
<td>I</td>
<td>LOXO-292</td>
<td>Dose finding</td>
<td>MTC, previously treated with vandetanib or cabozantinib</td>
<td>100% PD halted for toxicity</td>
<td>NCT0138642</td>
</tr>
<tr>
<td>II</td>
<td>Ponatinib</td>
<td>30 mg/day</td>
<td>FMTC</td>
<td>n.d.</td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>Ponatinib</td>
<td>n.a.</td>
<td>RET-mutated thyroid cancer</td>
<td>n.d.</td>
<td></td>
</tr>
<tr>
<td>II</td>
<td>Apatinib</td>
<td>500 mg/day</td>
<td>Thyroid cancer</td>
<td>n.d.</td>
<td>NCT0180542</td>
</tr>
<tr>
<td>II</td>
<td>Nintedanib</td>
<td>200 mg bid</td>
<td>MTC/DMTC</td>
<td>n.d.</td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>Dovitinib + paclitaxel</td>
<td>200 mg/day</td>
<td>Multi-histology</td>
<td>100% SD (RETmut non-MTC)</td>
<td>(94)</td>
</tr>
<tr>
<td>II</td>
<td>Pazopanib</td>
<td>800 mg/day</td>
<td>MTC</td>
<td>14% PR</td>
<td>(98)</td>
</tr>
<tr>
<td>I-Ib</td>
<td>RXDX-105</td>
<td>20–350 mg/day</td>
<td>BRAF- or RET-mutated cancer</td>
<td>1 PR</td>
<td>NCT01877811; (99)</td>
</tr>
<tr>
<td>I</td>
<td>BLU-667</td>
<td>Dose finding</td>
<td>MTC/DMTC/other RET-mutated cancer</td>
<td>n.d.</td>
<td></td>
</tr>
<tr>
<td>I</td>
<td>GSK3352589</td>
<td>1–100 mg/day</td>
<td>Healthy subjects</td>
<td>n.d.</td>
<td>NCT03154086</td>
</tr>
<tr>
<td>I</td>
<td>GSK3179106</td>
<td>10–200 mg/day</td>
<td>Healthy subjects</td>
<td>n.d.</td>
<td>NCT02798991</td>
</tr>
<tr>
<td>I</td>
<td>GSK3179106</td>
<td>5–100 mg/day</td>
<td>Healthy subjects</td>
<td>n.d.</td>
<td>NCT02727823</td>
</tr>
<tr>
<td>I/I</td>
<td>Vandetanib + bortezomib</td>
<td>Up to 300 mg qd</td>
<td>MTC</td>
<td>29% PR</td>
<td>NCT00923247</td>
</tr>
<tr>
<td>I/I</td>
<td>Semaxanib + paclitaxel</td>
<td>110 mg/m² on days 1, 15, 18, 22 and 25</td>
<td>Head and neck</td>
<td>100% SD</td>
<td>(141)</td>
</tr>
<tr>
<td>I</td>
<td>Vandetanib + everolimus</td>
<td>Starting 100 mg/day</td>
<td>Advanced cancers</td>
<td>n.d.</td>
<td>NCT01582191</td>
</tr>
</tbody>
</table>

*In combination studies, the data refer to the RET inhibitor; *refers to the MTC population only, when the study involved other cancer types (unless specified).

ATC, anaplastic thyroid carcinoma; bid, *bis in die* (twice daily); DTC, differentiated thyroid carcinoma; MTC, medullary thyroid carcinoma; ORR, overall response rate; PD, progressive disease; PR, partial response; SD, stable disease.

Table continues...

that RET mutation-positive patients had higher benefit than RET-negative ones, although the trend did not reach statistical significance due to the limited number of patients in the latter group. Adverse events (AEs) were observed in more than 30% of patients and included diarrhea, nausea, rash and hypertension. About half (49%) of the patients had a vandetanib-related increase of TSH levels and 8% of cases developed QTc prolongation. Five patients died because of serious side effects. Based on these results, the FDA (in 2011) and the EMA (in 2012) approved the use of vandetanib for the treatment of symptomatic or progressive medullary thyroid cancer in patients with unresectable locally advanced or metastatic disease. More recently, a phase I/II study aimed to assess the drug’s safety, tolerance, pharmacokinetic as well as the antitumor activity in a cohort of 10 adolescents and 6 children affected by MEN2B-associated MTC (Fox et al. 2013). All the patients but one expressed the M918T RET mutation. The tumor size decreased in all the 15 patients carrying the mutation. The overall objective response rate was 44% (7/15 patients; 95% confidence interval (CI): 21–73%) with a decrease in calcitonin level of 59% after one cycle of therapy. However, three children experienced progression after an initial PR.

An alternative to vandetanib is currently represented by cabozantinib (Cometriq, XL-184, Exelixis Inc), a potent inhibitor of MET, VEGFR2/KDR, RET, as well as other receptor tyrosine kinases, such as KIT, AXL and FLT3. In *in vitro* biochemical assays, cabozantinib displays inhibitory activity against both wild-type and mutated
M918T and Y791F RET forms associated with MTC (Yakes et al. 2011, Bentzien et al. 2013). Cabozantinib received FDA approval in 2012 and EMA approval in 2014 for the treatment of patients with progressive metastatic MTC. The activity of cabozantinib was firstly assessed in a phase I study on 87 patients with different solid tumors, including 37 MTC patients (Kurzrock et al. 2010). In the MTC subgroup 29% of cases achieved a PR and 41% had a SD for at least 6 months. The subsequent multicenter, randomized, placebo-controlled phase III EXAM study (Elisei et al. 2012) involved 330 patients with histologically confirmed unresectable, locally advanced or metastatic MTC that were randomly assigned 2:1 either to cabozantinib (140 mg/daily) or to placebo. Forty-eight percent patients harbored RET mutations. The PFS observed for the cabozantinib arm was significantly longer than placebo (11 vs 4 months, hazard ratio (HR) 0.28). PR was achieved in 28% of patients with a median duration of response of 14.7 months, while the median overall survival (OS) did not significantly differ in the two arms: 26.6 months under cabozantinib and 21.1 in the placebo group (P=0.24). In the subgroups harboring RET mutation, a PR was observed in 34% of cases, PFS was 13.9 months and the OS was 44.3 months (vs 18.9 months in the placebo arm, P=0.025). The most frequent AEs of grade 3 and 4 were diarrhea, hand-foot syndrome, fatigue and hypertension.

Investigational drugs

Several small molecules have been designed, developed or repurposed as RET kinase inhibitors during the past decade (reviewed in Phay & Shah 2010, Mologni 2011, Borrello et al. 2013, Mologni et al. 2017a,b). Some have already been approved for non-MEN2 thyroid cancer and are under investigation for MEN2, others are in advanced clinical development or have been clinically tested in a limited number of patients.

Sorafenib (Nexavar, Bay 43-9006, Bayer Healthcare Pharmaceuticals Inc) is an orally available VEGFR1–3, RET, BRAF and PDGFR inhibitor. Its use in differentiated thyroid carcinoma (DTC) has been approved by the FDA and the EMA in 2013 and 2014, respectively (Brose et al. 2014). Sorafenib activity in MTC was assessed in two prospective phase II studies (Lam et al. 2010, Ahmed et al. 2011). Lam and colleagues enrolled 21 MTC patients observing a PR in 6% of cases and SD in 88%. The PFS was 17.9 months. Similarly, Ahmed and colleagues studied a cohort of 15 MTC: 25% of cases achieved a PR, and the authors reported 100% OS at 12 months. Adverse events observed upon treatment with sorafenib in MTC patients, as well as in other thyroid cancer types include hand-foot syndrome, diarrhea, skin rash and fatigue. Larger cohorts will have to be evaluated before drawing conclusions on the efficacy of sorafenib in these patients.

Lenvatinib (Lenvima, E7080, Eisai Co Ltd) is an orally available multi-targeted TKI with reported activity against VEGFR 1–3, FGFR 1–4, PDGFR alpha, RET and KIT (Matsui et al. 2008, Bruheim et al. 2011, Wiegering et al. 2014). Lenvatinib has been approved by the EMA and the FDA for the treatment of patients with progressive radioiodine-refractory DTC recurrent or metastatic (Nair et al. 2015). The use of lenvatinib in MTC has been explored by Schlumberger and colleagues in a multicenter phase II study involving 59 patients treated with 24 mg/day in 28-day cycles (Schlumberger et al. 2016). The primary endpoint of the study was ORR, which was 36% (all PRs). Patients with or without prior VEGFR-targeted treatment responded similarly (35% vs 36% RR, respectively). Median PFS was 9 months and the achieved response was not affected by the RET mutational status.

Among the most advanced investigational compounds, sunitinib (Sutent, SU11248; Pfizer Ltd) has completed phase I and II trials in thyroid cancer (Dawson et al. 2008, Carr et al. 2010, Atallah et al. 2016, Bikas et al. 2016, Ravaud et al. 2017) and is currently undergoing specific evaluation in RET fusion-positive tumors (Bikas et al. 2016). Sunitinib was part of a large indolinone compound collection at SUGEN Inc (later acquired by Pfizer), which was initially developed to block angiogenic kinases belonging to the split tyrosine kinase domain family (VEGFR, FGFR, PDGFR) (Sun et al. 1998, 1999, 2000). Later, a few closely related compounds from this series were shown to inhibit RET at submicromolar concentrations in vitro and in vivo (Kim et al. 2006, Mologni et al. 2006, Chow & Eckhardt 2007, Jeong et al. 2011). Among these, SU11248 showed the best pharmacologic properties and progressed into clinical trials. Thus, although the published anti-RET preclinical data are scarce, and despite the broad multikinase activity of sunitinib, its safety profile is already well established from previous trials for other targets (it is approved for renal-cell carcinoma and imatinib-refractory GIST). This prompted clinical investigation in thyroid cancer patients. Overall, Sutent achieved 20–40% objective responses and 70–80% disease control rate (including SD) in the thyroid cancer patients’ population, which is in line with the currently approved drugs. Sunitinib was generally well tolerated in these patients, with mostly grade 1 or 2 treatment-related adverse events. The most frequent
toxicities observed in the different studies were fatigue, hand-foot syndrome, leukopenia and diarrhea, in some cases reaching grade 3–4 severity and leading to treatment discontinuation (about 10%) or dose reduction from the recommended 37.5 to 25 mg/die. One report described cardiac events in 14% of patients, half of whom had a severe episode (Ravaud et al. 2017). None of the other studies in thyroid cancer patients reported cardiovascular events; however, sunitinib had been watched for cardiovascular risk in patients with renal-cell carcinoma and gastrointestinal stromal tumors (Chu et al. 2007). A KIF5B-RET-positive non-small-cell lung cancer (NSCLC) case was reported with rapid response to sunitinib and clinical improvement after three days of treatment, in a heavily compromised patient (Wu et al. 2015). However, treatment was discontinued after 10 weeks for toxicity. Unfortunately, significant toxic effects seem relatively common among patients treated with sunitinib, which likely reflects its lack of selectivity. This may limit its clinical efficacy.

Motesanib (AMG-706, Amgen/Takeda) was originally developed by Amgen as an anti-angiogenic drug targeting VEGFR and c-Kit (Polverino et al. 2006). Activity was also noted in vitro against RET kinase in the nanomolar range, although with 1-log reduced potency compared to the primary targets. Thus, RET was a clinically interesting off target for motesanib. The compound was highly selective vs a number of other kinases. This may be due to its ability to make contact with small gatekeeper residues, which RET (V804), VEGFR (V916) and c-Kit (T670) have in common. On the other hand, this is a vulnerability, as motesanib is totally inactive against gatekeeper mutants, such as RET V804M (Mologni et al. 2013). In further preclinical studies, motesanib inhibited wildtype but not mutant RET C634W and M918T phosphorylation in cells. Consequently, proliferation of cells driven by mutant RET was not affected. However, in animal models, motesanib showed antitumor activity against xenografts carrying a M918T mutant RET (Coxon et al. 2012). This apparent contradiction is probably explained by anti-angiogenic effects of the drug. Clinical development of motesanib was later taken up by Takeda. In phase II studies, the drug showed modest activity in MTC (2% PR, 81% SD) as well as in DTC (14% PR, 67% SD) patients (Sherman et al. 2008, Schlumberger et al. 2009). Moreover, pharmacokinetic analyses indicated that motesanib trough concentrations were lower in MTC compared to DTC patients. The vast majority of patients (88%) experienced AEs, with a significant proportion (38%) of grade 3 AEs (Schlumberger et al. 2009). Currently, motesanib is mostly undergoing clinical evaluation as a VEGFR, PDGFR and c-Kit inhibitor (Raghav & Blumenschein 2011).

Fostamatinib is a multikinase inhibitor prodrug currently investigated as a SYK inhibitor (Bajpai 2009). Also in this case, the compound was inadvertently found to block RET kinase activity (Clemens et al. 2009). A recent multi-histology trial evaluated four thyroid cancer patients (papillary and follicular type), two of whom achieved SD as their best response (Park et al. 2013). In addition, one of the two patients with pheochromocytoma had durable SD. Although no MTC patient was enrolled, these data may encourage investigation of fostamatinib in MEN2 patients.

Alectinib (Alecensa, CH5424802, Roche/Chugui) is an anaplastic lymphoma kinase (ALK) inhibitor, approved for the treatment of advanced NSCLC refractory to crizotinib (Gadgeel et al. 2014). It was subsequently shown to be an effective RET inhibitor in vitro and in vivo (Kodama et al. 2014). Although alectinib is not quite as potent against RET as it is against ALK, the compound is active on most RET mutants tested and shows very good selectivity for RET vs VEGFR, which may be a competitive advantage over competing drugs. Alectinib will soon be tested in RET-rearranged NSCLC and RET-mutated thyroid cancer patients, in a non-randomized phase I/II study at Dana-Farber Cancer Institute (NCT03131206). A recent study reported objective responses in two of four RET-rearranged NSCLC patients (Lin et al. 2016a,b). From a clinical standpoint, alectinib has the advantage of already being well characterized. Whether it will show the same spectacular efficacy in RET-positive tumors as observed in ALK-driven disease remains to be ascertained.

Loxo Oncology recently developed a selective RET inhibitor named LOXO-292 with >100× selectivity vs KDR/VEGFR2, which compares favorably with cabozantinib and vandetanib on both wild-type and mutant RET (Brandhuber et al. 2016). The molecule shows good oral bioavailability and pharmacokinetics (PK) in animals, as well as high efficacy in RET-dependent tumor models, including xenografts and patient-derived tumor grafts. A multicenter phase I trial in patients with advanced RET fusion-positive NSCLC and MTC is currently recruiting participants (NCT03157128), to define the maximum tolerated dose (MTD).

Ponatinib (Iclusig, AP-24534, Ariad Pharmaceuticals) was developed to circumvent the highly intractable ABL gatekeeper mutant T315I in Philadelphia-positive chronic myeloid leukemia (Zhou et al. 2011, Cortes et al. 2012). It has been approved for the treatment of adult patients with T315I-positive CML and ALL or patients that have failed...
all other TKIs. Most like the V804M/L mutants of RET, substitution of a bulky isoleucine for wild-type threonine in ABL kinase renders the drug-binding site unfit to accommodate imatinib as well as second-generation ABL inhibitors (Quintas-Cardama & Cortes 2008). Through an innovative design, ponatinib was specifically made to overcome the steric impediment provided by the mutant residue, thus making this drug highly active against gatekeeper mutants. Indeed, when ponatinib was shown to potently inhibit RET (De Falco et al. 2013, Mologni et al. 2013), V804M/L mutants were shown to be effectively blocked. At the time of writing this manuscript, a phase II trial in advanced thyroid cancer previously treated with vandetanib and cabozantinib has been terminated prematurely (NCT01838642), and a new study was announced to open soon. Only 2 patients could be evaluated in the closed trial (both RET mutation positive), with no objective response (best response: PD). Other phase II, open-label trials in advanced NSCLC with RET translocations are currently recruiting patients (NCT01813734 and NCT01935336). Primary outcome will be overall response rate at 2 and 5 years, respectively. A new basket trial is recruiting patients with any refractory metastatic tumor carrying alterations in ponatinib targets, including RET (NCT02272998). As ponatinib is affected by significant systemic toxicity, which led to early termination of the first trial, its use may be confined to second or third lines of TKI therapy. Alternatively, dose reduction might be explored (Pinilla-Ibarz et al. 2013).

Apatinib is a potent VEGFR-2 inhibitor, which also targets PDGFR-β, c-Src, c-Kit and RET (Scott et al. 2015). Recently, it was shown to block invasion by RET fusion-positive lung cancer cells in vitro (Lin et al. 2016a,b). Since apatinib was already in advanced clinical phases of development (Li et al. 2016) as an anti-angiogenic drug, it is now under evaluation in metastatic refractory thyroid cancer, including MTC (NCT03199677) as well as in RET-positive NSCLC (NCT02540824).

Nintedanib (Vargatef, Ofev, BIBF 1120, Boehringer Ingelheim) is an anti-angiogenic multikinase inhibitor targeting VEGFRs, PDGFRs and FGFRs (Capdevila et al. 2014) used to treat idiopathic pulmonary fibrosis and lung cancer. In a recent retrospective analysis, the drug was reported to have achieved one complete response (CR) and one SD in two RET-rearranged NSCLC patients (Gautschi et al. 2017). Interestingly, according to the same study, ponatinib obtained only stabilization in two treated patients, while alectinib (n=2) failed. However, numbers are too small to draw any conclusions. Cabozantinib, vandetanib and sunitinib, for which at least 10 patients were evaluable, achieved 37, 18 and 22% overall responses, respectively (Gautschi et al. 2017), in line with observations in thyroid cancer. No data are available for the use of nintedanib in MEN2 or MTC, yet. However, a new phase II study is ongoing in MTC or DTC patients progressing after first-line therapy (NCT01788982).

Dovitinib (TKI258, CHIR258, Novartis) is another multi-targeted angiokinase inhibitor with anti-RET activity. It was investigated in a series of patients with various cancers (not MTC) and found to have sustained antitumor activity in two patients carrying a RET<sup>G691S</sup> germline variant (Quintela-Fandino et al. 2014). Interestingly, the same variant had previously been found in early-onset MEN2A patients and considered as a genetic modifier, cooperating with known oncogenic mutations to confer high penetrance an activating, oncogenic mutation (Robledo et al. 2003).

Pazopanib (Votrient, GW786034B, GlaxoSmithKline) is a VEGFR1/2/3, PDGFRα/β, and c-Kit inhibitor used for the treatment of renal-cell carcinoma (Sonpavde & Hutson 2007). It was tested in thyroid cancer patients on the basis of its anti-angiogenic properties, first in DTC (Bible et al. 2010) then in MTC (Bible et al. 2014). While in DTC patients the authors recorded 49% confirmed PRs, the results in MTC were less encouraging, with only 5/35 (14%) responding patients. One-third of patients had severe adverse events requiring treatment or dose reduction. Ignyta Inc is currently evaluating RXDX-105 (formerly CEP-32496) in a phase I clinical trial (NCT01877811) in patients with advanced solid tumors with RET or BRAF mutations or rearrangements (Patel et al. 2016). Although the trial is mostly intended for lung cancer patients, thyroid cancer patients are eligible to be enrolled as well. The compound was actually described as a poorly selective BRAF inhibitor, with potent off-target activity against RET and other kinases (James et al. 2012). Recently, its anti-RET activity was characterized in more detail in vitro and in vivo (Li et al. 2017). The same paper also reports a rapid PR in a NSCLC patient within the ongoing NCT01877811 trial. However, dose reduction was necessary due to toxicity. Indeed, broad inhibitory activity across the kinome of this compound may limit its efficacy in patients.

Next generation of RET targeted drugs

A new wave of RET-specific compounds is coming up in the next few years. By looking at recently filed patent applications, we get the feeling that the RET inhibitor field is about to burst. Likely, the new interest in RET
kinase inhibition has been fostered by the discovery of driver RET mutations in NSCLC and colorectal carcinoma. Although representing a minority of total lung and colon cancer patients (2% and 0.2%, respectively), RET fusion-positive cases are still a numerically important population of individuals (estimated 50,000 new cases per year worldwide), which adds up to the number of RET-positive thyroid cancer patients (about 100,000 per year). Moreover, the dismal prognosis of lung and colon cancers make new targeted treatments very attractive. In contrast, the great majority of thyroid cancers (papillary and follicular types) are characterized by high cure rates, while MTC is a rare disease. With cancer genome sequencing approaching routine use, it is probable that more RET-positive cancer types will be detected in the near future. Hence, RET has entered the stage of ‘interesting’ molecular targets in oncology. This has spurred a race toward RET-specific drug design programs, from which MEN2 patients will definitely benefit. Here we give a brief overview of forthcoming compounds, as of fall 2017.

Among new anti-RET drugs, BLU-667 of Blueprint Medicines has just started clinical phase I trial in 2017 (NCT03037385). It is claimed to be a potent and selective inhibitor of RET mutations and fusions, including drug-resistant mutants. The candidate was selected from a series of pyridinyl-pyrimidine derivatives, some of which showed low nanomolar activity against the gatekeeper V804L mutant in preclinical in vitro studies (Fleming et al. 2016, Rahal et al. 2016, 2017). The trial will evaluate safety and tolerability of BLU-667 in patients with RET-positive NSCLC and MTC.

GlaxoSmithKline developed two series of compounds specifically designed to inhibit RET kinase in the nanomolar range (Eidam et al. 2014, Cheung et al. 2016). Their clinical candidates (GSK3352589 and GSK3179106) have recently started phase I, first-in-human, dose escalating controlled studies to evaluate the safety, tolerability and pharmacokinetics in normal healthy volunteers (NCT03154086, NCT02798991 and NCT02727283). Although GSK seems to be developing these compounds for the treatment of irritable bowel syndrome (IBS), they may be as well effective for MEN2 and other RET-driven cancers (Abdel-Magid 2015).

Another interesting family of potential RET inhibitors is represented by quinazoline compounds described by Cancer Research Technology (Goldberg et al. 2015, Newton et al. 2016). Structurally related to vandetanib, these compounds are shown to possess striking selectivity (>100×) over VEGFR2/KDR kinase, which is a very common co-target of several RET inhibitors, whose inadvertent inhibition may potentially cause unwanted side effects and confounding anti-angiogenic activity. Therefore, these molecules may have improved therapeutic index compared to current drugs. The key to increased selectivity compared to vandetanib was a change in substituents around the phenyl ring, which also led to improved metabolic and pharmacokinetic features (Newton et al. 2016). This family was further elaborated in a more recent work, where the authors identified compounds with improved cellular selectivity (Jordan et al. 2016).

Similarly, a very recent patent filed by Array Biopharma claims potent nanomolar RET inhibitors with great selectivity (>30×) vs KDR kinase (Andrews et al. 2017). The inventors also propose the use of combinations including their lead compounds and at least one additional agent, to be chosen among known effective drugs for the treatment of cancer and IBS.

Nerviano Medical Sciences (NMS) has launched an ambitious program for the discovery of RET-selective inhibitors (Angiolini et al. 2014, Menichincheri et al. 2014). Two families of compounds were developed following a common concept of gatekeeper by-pass, shared with ponatinib (see above). In a first series, a triple bond linkage extends from a purine-based core scaffold buried in the active site through binding to the hinge region, bypassing the narrow space around a bulky gatekeeper such as V804M/L mutants of RET. Another recent patent filed by NMS describes compounds with low nanomolar activity and remarkable selectivity for RET+ cells in vitro. Again, the compounds are built by a similar design, with an extended linker joining two parts of the molecule, which are likely to fit nicely into the ATP pocket of the kinase even in the presence of large residues at the 804 position. The best compounds from this series are claimed to possess picomolar activity against MTC cells and are currently under preclinical development, with promising activity in RET-driven animal models (Ardini et al. 2017).

Dar and colleagues (University of California/Mount Sinai School of Medicine) recently described the discovery of potent RET inhibitors showing a high therapeutic window (Dar et al. 2012). Instead of using a classical biochemical screening based on the kinase activity, the authors exploited a Drosophila in vivo MEN2B model coupled with genetic and chemical profiling, toward a high-throughput simultaneous readout of target (RET) and anti-target inhibition. This strategy allowed the identification of hit compounds (AD80 and AD81) with the desired activity and devoid of unwanted toxicity. The findings translated into improved in vivo efficacy and
tolerability in a mouse xenograft model. Whether this will also hold in humans remains to be demonstrated.

NVP-AST487 is a Flt-3, VEGFR, PDGFR, c-Kit and c-Abl inhibitor with nanomolar activity against RET C634W-transformed NIH3T3 xenografts (Åkeno-Stuart et al. 2007) and in MEN2-derived human cell lines (Gild et al. 2013). It has recently been used to target GDNF-stimulated wild-type RET receptor in MCF7 breast cancer cells, where a role for RET activation in resistance to aromatase inhibitors has been demonstrated (Andreucci et al. 2016).

Pz-1 is a very potent compound described by Synactix Pharmaceuticals (Frett et al. 2015), which was able to completely suppress RET-driven tumor growth in mice at 1 mg/kg. However, the compound also greatly reduced Ras-induced tumors, which raises doubts on its selectivity and therapeutic applicability. Also, strong VEGFR2 inhibition, as said, may help antitumor activity or cause adverse effects.

To conclude this section, it is worth mentioning that several academic groups, including our lab, have published interesting early phase medicinal chemistry papers describing the discovery of novel RET inhibitors with promising in vitro data, in the past three years. Often, efforts were focused on activity against the gatekeeper mutants (Dunna et al. 2015, Ferreira et al. 2015, Moccia et al. 2015, Han et al. 2016, Song et al. 2016, Yoon et al. 2016, 2017, Mologni et al. 2017a, b, Wang et al. 2017). Finally, in a completely different approach, Kumarasamy and Sun described the selective block of RET transcription by a G-quadruplex-stabilizing agent (Kumarasamy & Sun 2017). By hitting the promoter of RET gene, this strategy would result in suppression of all mutants, but it would not be useful for RET fusions. Similarly, antibody-based approaches such as one described by Takeda (Arai et al. 2007) would hit RET mutants expressed on the cell surface, but not fusion oncogenes, that are localized within the cytoplasm. More universal targeting approaches are possible, including antisense, RNAi, or ribozyme-based strategies (Parthasarathy et al. 1999, Backman et al. 2003). However, all nucleic acid-based therapeutics suffer from poor delivery.

**Combination therapies**

While targeted cancer monotherapy has demonstrated good efficacy when rationally designed, the combination of two or more agents may further improve the outcome, in an additive or synergistic manner, by causing a more profound anti-tumoral effect. More importantly, combination therapies have the potential of preventing drug resistance, which very often arises from the selection of pre-existing mutant subclones or from adaptive response to treatment (Gorre et al. 2001, Doelebe et al. 2012, Straussman et al. 2012). A few preclinical studies have addressed the issue of combined treatment in MEN2 disease and/or thyroid neoplasia. Lopergolo and colleagues at the National Cancer Institute in Milan, Italy, showed that addition of cisplatin synergistically improved antitumor activity of sunitinib in MTC xenografts, by potentiating the activation of CD95-mediated apoptosis (Lopergolo et al. 2014). In another interesting study, sorafenib was shown to cause only transient MAPK pathway inhibition in MTC cell lines at low doses. This led the authors to test whether its effects could be enhanced by concomitant treatment with a MEK inhibitor. Indeed, combination with selumetinib (AZD6244, AstraZeneca) greatly increased growth inhibition compared to either single agent (Koh et al. 2012). While this combination has not yet been further investigated in thyroid cancer, the addition of MEK/ERK inhibitors to upstream kinase inhibitors (e.g., BRAF) has proven very successful in other tumors (Robert et al. 2015).

The concept of hitting driver oncogenic pathways at multiple nodes was explored by Jin and coworkers (Jin et al. 2011). The authors initially sought to block MEK/ERK and PI3K/AKT/mTOR pathways in DTCs driven by RAS, RAF or PTEN mutations. To this end, they combined the dual PI3K/mTOR inhibitor NVP-BEZ235 (Novartis) with the pan-RAF inhibitor RAF265/CHIR-265 (Novartis), which targets VEGFR2 as well. However, they noted that RAF265 is structurally related to sorafenib; hence, it may also block RET kinase. Indeed, the drug was shown to be a potent RET inhibitor and cooperated with BEZ235 to inhibit TT cells (carrying a RET C634W allele) growth in vivo. It is possible that VEGFR2 targeting contributed to in vivo tumor growth inhibition, similarly to all other effective RET inhibitors. More recently, another group explored synergistic inhibition of RET-mediated signaling and cell growth by the combination of RAF265 with a different PI3K inhibitor, named ZSTK474 (Bertazza et al. 2015). ERK and AKT activation, cell growth and survival, and calcitonin expression and secretion were all significantly better inhibited by the combination compared to single treatments, in TT cells.

Another possible strategy may involve the use of heat shock protein inhibitors. As mentioned earlier, pharmacological inhibition of HSP90 by 17-AAG or ganetespib downregulates RET levels in cells (Alfano et al. 2010, Lin et al. 2017) and sorafenib induces a similar effect after prolonged treatment (Plaza-Menacho et al. 2007).
The combination of HSP90 and RET inhibitors may lead to improved block of oncogenic signaling in RET-addicted cells.

A few papers described the use of imatinib (Gleevec, STI-571, Novartis) against RET. However, the drug showed poor activity in preclinical and clinical studies (Cohen et al. 2002, de Groot et al. 2007). In one attempt to improve its efficacy in MTC models, authors combined imatinib with a FGFR4 inhibitor (PD173074, Ezzat et al. 2005). Although the drugs concentrations used in vitro were rather high, hardly achievable in vivo, the combination did obtain better inhibition of TT xenografts, at reasonable doses.

On the clinical side, a phase I/II trial of vandetanib plus the proteasome inhibitor bortezomib (Velcade, Millennium Pharmaceuticals) was run in adults with hereditary or sporadic, locally advanced or metastatic MTC. Phase I enrolled 21 patients to find the MTD. Analysis of tumor response in this group showed 29% PR (6/21), while only one patient was enrolled in the phase II and progressed (NCT00923247). A possibly interesting activity of semaxanib (SU5416) in combination with paclitaxel in two metastatic MTC patients remained anecdotal because the compound has then been discontinued. The trial was actually designed for head-and-neck cancers, including thyroid neoplasm, and semaxanib was meant as an anti-angiogenic drug. Interestingly, the only two MTC patients enrolled were among the four patients (out of 11 evaluable) showing prolonged SD (Cooney et al. 2005). The documented anti-RET activity of the compound (Mologni et al. 2006) may explain these results. A recent single-arm phase 1 trial is currently recruiting patients with advanced cancers, to be treated with a combination of vandetanib plus everolimus, an mTOR inhibitor (NCT01582191). The idea is based on everolimus activity against ABC transporters, which limit drug efficacy, especially in the central nervous system. Indeed, a first successful case has been reported (Subbiah et al. 2015). Whether these results will be relevant to the MEN2/MTC population remains to be seen.

Finally, an innovative precision therapy trial (NCT02363647) is inviting patients with sporadic metastatic MTC. After next-generation genetic analysis to identify tumor drivers, a Drosophila model is rapidly generated and tested against a panel of FDA-approved drugs or drug combinations, to identify and propose a possible personalized treatment. Time will tell if this strategy will obtain superior response or cure rates compared to current approaches.

Conclusions and future hope

The discovery of RET mutations in MEN2 syndrome 25 years ago opened a new era for the management of the disease. After two decades of intense research, potent RET inhibitors have reached the clinical scene, with many more to come in the next few years (Mologni et al. 2017a,b). As physicians know very well, to have a full array of drugs to treat cancer is a most desirable situation, when we are confronted with an enemy that changes rapidly to survive our attacks. However, it is currently unclear what are the mechanisms of primary or acquired resistance to RET inhibitors in thyroid cancer. This information is needed in order to rationally devise effective alternative targeted treatments, as demonstrated for CML patients (Redaelli et al. 2012). Surely, RET gatekeeper mutations cause a sharp decrease of efficacy for some drugs in vitro (e.g. vandetanib, Carlomagno et al. 2004). A recent work reported NRAS mutations in ponatinib-resistant RET-rearranged NSCLC cells (Nelson-Taylor et al. 2017). It is also unknown whether sequential targeted therapies with multiple RET inhibitors will improve patients OS. Finally, all current drugs are anti-angiogenic compounds with limited selectivity for RET. Therefore, the relative contribution of anti-RET and anti-VEGFR activities in determining the clinical outcome is not known, although the ZETA trial indicated that RET-positive patients may respond better, suggesting a specific anticancer role for RET inhibition (Wells et al. 2012).

It has been an exciting time from the discovery of the target to the development of targeted treatments. Much remains to be done, but we (and Mary) can look forward with new optimism.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of this review.

Funding

This work did not receive any specific grant from any funding agency in the public, commercial or not-for-profit sector.

References

Abdel-Magid AF 2015 RET kinase inhibitors may treat cancer and gastrointestinal disorders. ACS Medicinal Chemistry Letters 6 13–14. (https://doi.org/10.1021/ml500402t)

Cohen MS, Hussain HB & Moley JF 2002 Inhibition of medullary thyroid carcinoma cell proliferation and RET phosphorylation by tyrosine kinase inhibitors. Surgery 132 960-966; discussion 966–977. (https://doi.org/10.1016/s0039-6060(01)01285-2)

Cooney MM, Tserng KY, Makar V, McPeak RJ, Ingalls ST, Dowlati A, Cohen MS, Hussain HB & Moley JF 2002 Inhibition of medullary thyroid carcinoma cell proliferation and RET phosphorylation by tyrosine kinase inhibitors. Surgery 132 960-966; discussion 966–977. (https://doi.org/10.1016/s0039-6060(01)01285-2)


Dawson SJ, Conus NM, Toner GC, Raleigh JM, Hicks RJ, McArthur G & Rischin D 2008 Sustained clinical responses to tyrosine kinase inhibitor sunitinib in thyroid carcinoma. Anticancer Drugs 19 547–552. (https://doi.org/10.1097/01.CAD.0b013e328286c6d7)


Gild ML, Landa I, Ryder M, Ghoose RA, Knauf JA & Fagin JA 2013 Targeting mTOR in RET mutant medullary and differentiated thyroid cancer cells. Endocrine-Related Cancer 20 659–667. (https://doi.org/10.1530/ERC-13-0085)


Han M, Li S, Ai J, Sheng R, Hu Y & Geng M 2016 Discovery of 4-chloro-3-(5-(pyridin-3-yl)-1,2,4-oxadiazole-3-yl)benzamides as novel RET


Mars JX 2005 Molecular genetics of some endocrine neoplasias type 1 and 2. Nature Reviews Cancer 5 367–375. (https://doi.org/10.1038/nrc1610)


New treatments for MEN2


Yuan ZL, Guan YJ, Yang JC 2004 Rapid response to sunitinib in a patient with lung adenocarcinoma harboring KIF5B-RET fusion gene. *Journal of Thoracic Oncology*** 10 e95–e96. (https://doi.org/10.1097/JTO.0000000000000611)

Received in final form 7 November 2017
Accepted 14 November 2017