Therapeutic Potential of PI3K/AKT/mTOR Pathway

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Gastrointestinal stromal tumor (GIST) originates from interstitial cells of Cajal (ICCs) in the myenteric plexus of the gastrointestinal tract. Most GISTs arise due to mutations of KIT and PDGFRA gene activation, encoding the receptor tyrosine kinase (RTK). The clinical use of the RTK inhibitor imatinib has significantly improved the management of GIST patients; however, imatinib resistance remains a challenge. The phosphatidylinositol 3-kinase (PI3K)/protein kinase B (AKT)/mammalian target of rapamycin (mTOR) pathway is a critical survival pathway for cell proliferation, apoptosis, autophagy and translation in neoplasms. Constitutive autophosphorylation of RTKs has an impact on the activation of the PI3K/AKT/mTOR pathway. In several preclinical and early-stage clinical trials PI3K/AKT/mTOR signaling inhibition has been considered as a promising targeted therapy strategy for GISTs. Various inhibitory drugs targeting different parts of the PI3K/AKT/mTOR pathway are currently being investigated in phase I and phase II clinical trials. This review highlights the progress for PI3K/AKT/mTOR-dependent mechanisms in GISTs, and explores the relationship between mTOR downstream signals, in particular, eukaryotic initiation factors (eIFs) and the development of GISTs, which may be instrumental for identifying novel therapeutic targets.

Keywords: gastrointestinal stromal tumors ; PI3K/AKT/mTOR ; inhibitor ; eIFs

1 The PI3K/AKT/mTOR Pathway in GISTs

1.1. Influence on Cell Proliferation

It is well established that mTOR can assist the integration of nutrient and mitogen signals into intracellular, thereby regulating cell growth and cell division; meanwhile, rapamycin, an mTOR inhibitor, inhibits cell growth and cell cycle [1]. The overexpression of constitutively active mutated S6K1 or wt eIF4E accelerated G1-phase progression, suggesting that S6K1 and 4E-BP1/eIF4E as downstream signals of mTOR, regulates cell proliferation to a certain extent by controlling the cell cycle [2]. In a study of 108 GIST patients, the phosphorylation of p70S6K and 4E-BP1 was separately detected in *KIT*-mutated GISTs at 38%, in *PDGFRA*-mutated GISTs at 83%, and in wt GISTs at 74% [3]. Interestingly, mTOR was overexpressed in *PDGFRA*-mutated and wt GISTs, indicating that mTOR-related inhibitors may have therapeutic effects on primary imatinib-resistant GISTs [3]. Pang and colleagues demonstrated that mTORC1 signaling was inactivated by DEPDC5, with the suppression of the phosphorylation of p70S6K and S6, resulting in reduced cell proliferation and subsequently cell-cycle arrest in GIST cells [4]. Li and colleagues showed that PI3K/AKT/mTOR signaling was inactivated due to *FANS* knockdown, especially with the attenuation of the activation of RPS6 and 4E-BP1, leading to the inhibition of proliferation and migration in GIST cells [5].

1.2. Influence on Apoptosis

As the first study linking apoptosis to prognosis of GIST patients, Wang and colleagues found in 2007 that the apoptotic index was gradually decreased in tumor tissue specimens of patients with GISTs. The authors suggested that programmed cell death may be avoided in the pathogenesis of GISTs [6]. More recently, PI3K/AKT/mTOR-regulated apoptosis has been widely investigated in GISTs. Activated PI3K can directly inhibit apoptosis in tumors [7]. AKT, an anti-apoptotic factor, mediates these PI3K-dependent cell survival responses [8]. In GISTs, the activation mutation of the upstream c-KIT and PDGFRA is the initial mechanism of the PI3K/AKT/mTOR signal activation [83]. Ma and colleagues demonstrated KIT expression as positively correlating with the cell proliferation marker Ki-67 and a converse correlation with the pro-apoptotic protein APAF. Thereby *c-KIT* participates in GIST tumorigenesis by inducing proliferation and reducing apoptosis [9]. Ihle and colleagues reported that the downregulation of miR-221 and miR-222 induced apoptosis via the KIT/AKT pathway in GIST cells; the KIT/AKT pathway has effects on tumor progression, controlling cell proliferation and apoptosis [10].

1.3. Influence on Autophagy

Autophagy is a highly conserved catabolic process that participates in cell survival and in preserving cell metabolic balance [11][12]. In various tumors, autophagy is critically controlled by the PI3K/AKT/mTOR pathway [13]. Via this pathway, increased expression levels of autophagy related genes may enhance metastatic spread in paranasal squamous cell carcinomas [14]. Autophagy can promote cell adaption and survival, but it also leads to cell death under particular conditions. Autophagy is a double-edged sword for drug resistance, as growing evidence indicates that autophagy can contribute to resistance against chemotherapeutics [15]. In recent years, it has become evident that autophagy might be critical in GIST progression. Beclin1 (BECN1) forms the BECN1–PIK3C3–PIK3R4 complex by aggregating cofactors, at the same time the autophagy protein cascade is activated [16]. Miselli and colleagues demonstrated that GIST patients responded to imatinib treatment by autophagy instead of apoptosis [17]. High levels of pro-autophagy beclin1/PI3K III and low levels of anti-autophagy beclin1/bcl2 complexes are consistent with the existence of autophagy in imatinib-treated GIST patients; thus suggesting autophagy playing a role in the underlying molecular mechanism of how GISTs form [17]. Wei and colleagues reported that beclin-1 knockdown significantly enhanced the sensitivity of GIST cells to imatinib; while miR-30a, directly targeting *Beclin1* also enhanced imatinib sensitivity through the downregulation of *Beclin1* [18]. Upon treatment, the sizeable GIST cell subpopulations survive and remain quiescent for a long time, leading to acquired resistance and treatment failure. Gupta and colleagues confirmed that a considerable number of GIST cells enter a reversible resting state by activating autophagy-dependent survival mechanisms after imatinib treatment [19]. GIST cells were destroyed by the synergistic effect of imatinib and autophagy inhibition by RNAi-mediated silencing against ATG7 and ATG12 [12]. The standard first-line molecular-targeted therapy in GISTs is focused on imatinib for GISTs; but primary or secondary resistance is becoming increasingly prominent. Fortunately, some studies have focused on new molecular mechanisms, such as autophagy, in order to address drug resistance. Hsueh and colleagues reported that NVP-AUY922, an HSP90AA1 inhibitor, of which KIT is a client, regulated autophagy-mediated pathways to downregulate the expression of the KIT protein and to inhibit GIST cell growth [93]. Rapamycin, an mTOR inhibitor and autophagy inducer, enhanced autophagy activity, downregulated the expression of KIT, and led to apoptosis in GIST430 and GIST48 cell lines [20].

2. eIFs and PI3K/AKT/mTOR Pathway

2.1. The Relationship between eIFs and the PI3K/AKT/mTOR Pathway

Protein translation has committed effects on the process of eukaryotic gene expression [21][22]. The translation includes the following four steps: initiation, elongation, termination, recycling. It is well established that the rate-limiting step is the initiation phase regulated by eukaryotic translation initiation factors (eIFs) [21][23]. The initiation step starts with the assembly of an elongation-appropriate complex, of a 43S pre-initiation complex that is composed of the 40S small ribosomal subunit, methionine tRNAi, and a group of eIFs. Subsequently, the 43S pre-initiation complex binds to the 5’ end of mRNA and then to the eIF4F complex that mediates the recruitment of ribosomes to Mrna [23][24]. The eIF4F complex consists of three polypeptides (eIF4E, eIF4A and eIF4G). eIF4E is a cap-binding protein that recognizes and binds to the mRNA 5' m^7G cap structure, which interacts with the DEAD box RNA helicase eIF4A and the scaffolding protein eIF4G. The function of eIF4G is to bridge the mRNA with the ribosome, enhance the helicase activity of eIF4A, and realize the mRNA circularization [25]. Through the PI3K/AKT/mTOR pathway, extracellular stimuli induce phosphorylation and the inactivation of the 4Ebinding proteins (4E-BP1, 2 and 3) that influence eIF4F activity [24] . Thus, the high influence of eIFs on protein translation is of utmost interest in targeted cancer therapies.

In a review about the coverage of the mTOR pathway by Next-Generation Sequencing (NGS) oncology panels, eIFs, a group of mTOR downstream proteins, were reported with low mutational frequencies of a rate less than 1% and no reported drug sensitivity alterations [26]. Numerous marker genes are known to have great impact on disease progression and prognosis, even though they are rarely mutated. eIF subunits are important examples of these markers [26].

2.2. PI3K/AKT/mTOR-Regulated eIFs as a Potential Therapeutic Target in Tumors

eIFs may become promising therapeutic targets for many tumors under the PI3K/AKT/mTOR pathway control. The eIF4F complex is a critical part under the regulation of this signaling axis [25][27][28]. In Abl-expressing leukemic cells, eIF4B stimulated eIF4F activity by increasing the eIF4A RNA helicase activity on the mRNA 5′UTR, integrating signals from the PI3K/AKT/mTOR pathway [29]. When those inhibitors of PI3K (with LY294002), AKT (with Akti-1/2) or mTOR (with rapamycin) were separately applied to Abl-transformed cells, the phosphorylation of eIF4B Ser422 markedly decreased, which inhibited cell proliferation and growth [29]. Cencic and colleagues highlighted that eIF4A reversed drug resistance by curtailing the potential activity of translation initiation in lymphomas [30]. Hippuristanol, a translation initiation inhibitor,

specifically inhibits eIF4A. Hippuristanol plus ABT-737 (an inhibitor of *Bcl-2)* synergistically increased cell death in Myc-driven lymphomas; likewise, the inhibition of eIF4AI is enough to improve the chemosensitivity of Myc-driven lymphomas to ABT-737 [30].

Furthermore, some studies focused on the PI3K/AKT/mTOR signaling members and other eIFs in human tumors. Golob-Schwarzl and colleagues demonstrated that eIF5 has the potential as a biomarker to indicate whether there is virus infection in hepatocellular carcinoma (HCC) tissue [31]. eIF5 was downregulated in non-virus related HCC; nevertheless, it revealed an overexpression in HBV-associated HCC. The downregulation of p-mTOR and mTOR was observed in HBV-associated HCC. The expression of other eIFs, such as eIF2a, eIF3D, eIF3H, eIF3I, eIF3J, eIF4E and eIF6 was downregulated in HBV-associated HCC [31]. Tapia and colleagues investigated the activation of PI3K/AKT/mTOR signals in gastric cancer (GC), with the overexpression of most important target proteins of the pathway, such as PI3K, AKT, p-AKT, p-mTOR, p-4E-BP1, P70S6K1, p-P70S6K1, eIF-4E, and p-eIF-4E proteins in tumor tissue [32]. Low expression of 4E-BP1 has a poor overall survival, which points towards a potential role as prognostic marker [32]. Wang and colleagues demonstrated EIF3B expression as upregulated in advanced GC patients who have a short 5-year survival [33]. In vitro and in vivo studies into the downregulation of EIF3B showed the inhibition cell proliferation and clonogenicity in SGC7901 and BGC823 cells lines, and knockdown of EIF3B notably abated the tumor volume and weight in an SGC7901 xenograft mouse model. Interestingly, the PI3K/AKT/mTOR signaling activity is related with an upregulation of eIF3B [33]. Golob-Schwarzl and colleagues showed the expression of eIFs family members, bringing eIF1, eIF5, and eIF6 to attention, together with components of the PI3K/AKT/mTOR signaling cascade in colorectal cancer (CRC) [34]. These eIF subunits and the PI3K/AKT/mTOR signaling members had a significant influence on the overall survival of CRC patients [34].

In summary, the PI3K/AKT/mTOR pathway has important effects on the basic intracellular functions, including cell growth, apoptosis, translation, and cell metabolism [35]. Especially, the regulation of translation is critical for maintaining homeostasis in cells [21]. The dysregulation of protein synthesis may be related to the abnormal expression of eIFs and altered activation of the PI3K/AKT/mTOR pathway [24][26].

3. Challenges and Future Directions

More than 80% of patients with advanced GISTs have remarkable clinical benefits from molecular targeted therapy, but secondary resistance is found in half of these GIST patients [36]. Imatinib as the first-line therapy effectively provides long-term disease stability and prolongs the overall survival in *KIT*-mutated GIST patients [36][37]. In a multicenter clinical study, only 10% of imatinib-treated patients had a 10-year progression-free survival, and interestingly GIST patients with *KIT exon 11* mutations suggest a better outcome beyond 10 years [38]. GIST patients with *KIT exon 9* mutations had a significantly worse prognosis compared to those with *KIT exon 11* mutations [37], who also exhibited worse response to imatinib treatment [39]. Other TKIs, such as sunitinib and regorafenib, have complementary activity in that sunitinib targets the ATP-binding pocket to inhibit imatinib-resistance mutations, whereas regorafenib primarily targets the activation loop to achieve similar effects [40]. Once imatinib resistance emerges, sunitinib and regorafenib as second- and third-line molecular targeted therapies, could provide clinical benefit in a short-term [41][42][43]. For imatinib-resistant GIST patients, the tumorigenesis mainly involved in the heterogeneity of *KIT* secondary mutations [44], and targeting *KIT,* is limited by its susceptibility to multiple mutations [39]. It may be a key to inverse secondary resistance in GISTs through selecting agents with known pre-clinical activity for mutations that form the basis for resistance, or that overcome the effect of resistance mutations.

The PI3K/AKT/mTOR signaling axis is a crucial survival pathway in imatinib-resistant GISTs [45][46]. Subsequently, many studies paid attention on directly inhibiting downstream of this pathway in GISTs, in order to interrupt more comprehensive RTK signals as much as possible. Therefore, more research will have to pay attention to the potential of those inhibitors of PI3K (BKM120, BYL719, GDC-0941), AKT (Perifosine), mTOR (RAD001) and Dual PI3K/mTOR (BEZ2335, GDC-0980), combined with imatinib. Parts of combination treatment strategies are being investigated in early-stage clinical studies. For instance, BKM120 plus imatinib in combination could inhibit cell growth in imatinib-resistant GIST cell lines, whereas imatinb combined with GDC-0941 or with BEZ2335 improved the efficacy with a more pronounced tumor volume reduction in all GIST xenograft models. In addition, mTOR activity is related to the mutation type status. The mTOR pathway is activated in *PDGFRA* mutated and wt GISTs, which suggests that mTOR or upstream mTOR inhibitors may be potential therapeutic targets for primary resistant GISTs [3]. In mouse models, pharmacologic inhibitors of the PI3K pathway diminished tumor proliferation in tumor-bearing single-mutated $KIT^{V558\Delta/+}$ mice, indicating that PI3K kinase makes a major contribution to tumor cell proliferation in established GISTs [47]. Thus, the interruption of the PI3K/AKT/mTOR pathway may represent a rational therapeutic approach in GIST patients.

Interestingly, the PI3K/AKT/mTOR pathway plays a pivotal role in mRNA translation, especially in the initiation phase. mTOR regulates the assembly of eIF4f to manage translation initiation. The PI3K/AKT/mTOR pathway affects tumorigenesis through eIFs, reported in HCC [31], GC [33], CRC [34], and other cancer types [29][48]. In summary, eIFs integrate the signals from the PI3K/AKT/mTOR pathway, which may be a promising target for tumor therapy in future. Regrettably, there is no literature on eIFs involved in GISTs. Based on the above principles, the hypothesis, that eIFs may represent a therapeutic target in GISTs via the PI3K/AKT/mTOR pathway, merits further investigation.

4. Conclusion

Although most patients with advanced GISTs display remarkable clinical benefits from TKI therapy by imatinib, resistance to TKIs occurs in half of those GIST patients. Further therapeutic approaches are urgently needed. The PI3K/AKT/mTOR pathway is not only vital in tumorigenesis, but also important for novel potential therapeutic targets. To date, several inhibitors targeting different parts of the PI3K/AKT/mTOR pathway have been investigated in imatinib-resistant GISTs, including PI3K inhibitors, AKT inhibitors, mTOR inhibitors and dual PI3K/mTOR inhibitors. These drugs combined with imatinib may potentially provide a broader inhibition of pro-growth cellular mechanisms than the single-agent imatinib. Besides, the PI3K/AKT/mTOR pathway has a pivotal effect on mRNA translation initiation, directly regulated by eIFs that may also be a promising future therapeutic target for these tumors.

References

1. Teng, Q.X.; Ashar, Y.V.; Gupta, P.; Gadee, E.; Fan, Y.F.; Reznik, S.E.; Wurpel, J.N.D.; Chen, Z.S. Revisiting mTOR inhibitors as anticancer agents. Drug Discov. Today 2019, 24, 2086–2095, doi:10.1016/j.drudis.2019.05.030.
2. Fingar, D.C.; Richardson, C.J.; Tee, A.R.; Cheatham, L.; Tsou, C.; Blenis, J. mTOR controls cell cycle progression through its cell growth effectors S6K1 and 4E-BP1/eukaryotic translation initiation factor 4E. Mol. Cell. Biol. 2004, 24, 200–216, doi:10.1128/mcb.24.1.200-216.2004.
3. Sápi, Z.; Füle, T.; Hajdu, M.; Matolcsy, A.; Moskovszky, L.; Márk, A.; Sebestyén, A.; Bodoky, G. The activated targets of mTOR signaling pathway are characteristic for PDGFRA mutant and wild-type rather than KIT mutant GISTs. Diagn. Mol. Pathol. 2011, 20, 22–33, doi:10.1097/PDM.0b013e3181eb931b.
4. Pang, Y.; Xie, F.; Cao, H.; Wang, C.; Zhu, M.; Liu, X.; Lu, X.; Huang, T.; Shen, Y.; Li, K.; et al. Mutational inactivation of mTORC1 repressor gene DEPDC5 in human gastrointestinal stromal tumors. Proc. Natl. Acad. Sci. USA 2019, 116, 22746–22753, doi:10.1073/pnas.1914542116.
5. Li, C.F.; Fang, F.M.; Chen, Y.Y.; Liu, T.T.; Chan, T.C.; Yu, S.C.; Chen, L.T.; Huang, H.Y. Overexpressed Fatty Acid Synthase in Gastrointestinal Stromal Tumors: Targeting a Progression-Associated Metabolic Driver Enhances the Antitumor Effect of Imatinib. Clin. Cancer Res. 2017, 23, 4908–4918, doi:10.1158/1078-0432.CCR-16-2770.
6. Wang, Q.; Kou, Y.W. Study of the expressions of p53 and bcl-2 genes, the telomerase activity and apoptosis in GIST patients. World J. Gastroenterol. 2007, 13, 2626–2628, doi:10.3748/wjg.v13.i18.2626.
7. Wendel, H.G.; De Stanchina, E.; Fridman, J.S.; Malina, A.; Ray, S.; Kogan, S.; Cordon-Cardo, C.; Pelletier, J.; Lowe, S.W. Survival signalling by Akt and eIF4E in oncogenesis and cancer therapy. Nature 2004, 428, 332–337, doi:10.1038/nature02369.
8. Hennessy, B.T.; Smith, D.L.; Ram, P.T.; Lu, Y.; Mills, G.B. Exploiting the PI3K/AKT pathway for cancer drug discovery. Nat. Rev. Drug Discov. 2005, 4, 988–1004, doi:10.1038/nrd1902.
9. Ma, Y.Y.; Yu, S.; He, X.J.; Xu, Y.; Wu, F.; Xia, Y.J.; Guo, K.; Wang, H.J.; Ye, Z.Y.; Zhang, W.; et al. Involvement of c-KIT mutation in the development of gastrointestinal stromal tumors through proliferation promotion and apoptosis inhibition. OncoTargets Ther. 2014, 7, 637–643, doi:10.2147/OTT.S60458.
10. Ihle, M.A.; Trautmann, M.; Kuenstlinger, H.; Huss, S.; Heydt, C.; Fassunke, J.; Wardelmann, E.; Bauer, S.; Schildhaus, H.U.; Buettner, R.; et al. miRNA-221 and miRNA-222 induce apoptosis via the KIT/AKT signalling pathway in gastrointestinal stromal tumours. Mol. Oncol. 2015, 9, 1421–1433, doi:10.1016/j.molonc.2015.03.013.
11. Scott, R.C.; Juhász, G.; Neufeld, T.P. Direct induction of autophagy by Atg1 inhibits cell growth and induces apoptotic cell death. Curr. Biol. 2007, 17, 1–11, doi:10.1016/j.cub.2006.10.053.
12. Lee, S.B.; Kim, S.; Lee, J.; Park, J.; Lee, G.; Kim, Y.; Kim, J.M.; Chung, J. ATG1, an autophagy regulator, inhibits cell growth by negatively regulating S6 kinase. EMBO Rep. 2007, 8, 360–365, doi:10.1038/sj.embor.7400917.
13. Kondo, Y.; Kanzawa, T.; Sawaya, R.; Kondo, S. The role of autophagy in cancer development and response to therapy. Nat. Rev. Cancer 2005, 5, 726–734, doi:10.1038/nrc1692.

14. Biray Avci, C.; Sezgin, B.; Goker Bagca, B.; Karci, H.B.; Gode, S. PI3K/AKT/mTOR pathway and autophagy regulator genes in paranasal squamous cell carcinoma metastasis. Mol. Biol. Rep. 2020, 47, 3641–3651, doi:10.1007/s11033-020-05458-8.

15. Xu, Z.; Han, X.; Ou, D.; Liu, T.; Li, Z.; Jiang, G.; Liu, J.; Zhang, J. Targeting PI3K/AKT/mTOR-mediated autophagy for tumor therapy. Appl. Microbiol. Biotechnol. 2020, 104, 575–587, doi:10.1007/s00253-019-10257-8.

16. Han, T.; Guo, M.; Gan, M.; Yu, B.; Tian, X.; Wang, J.B. TRIM59 regulates autophagy through modulating both the transcription and the ubiquitination of BECN1. Autophagy 2018, 14, 2035–2048, doi:10.1080/15548627.2018.1491493.

17. Miselli, F.; Negri, T.; Gronchi, A.; Losa, M.; Conca, E.; Brich, S.; Fumagalli, E.; Fiore, M.; Casali, P.G.; Pierotti, M.A.; et al. Is autophagy rather than apoptosis the regression driver in imatinib-treated gastrointestinal stromal tumors? Transl. Oncol. 2008, 1, 177–186, doi:10.1593/tlo.08157. PMID: 19043528.

18. Chen, W.; Li, Z.; Liu, H.; Jiang, S.; Wang, G.; Sun, L.; Li, J.; Wang, X.; Yu, S.; Huang, J.; et al. MicroRNA-30a targets BECLIN-1 to inactivate autophagy and sensitizes gastrointestinal stromal tumor cells to imatinib. Cell Death Dis. 2020, 11, 1–13, doi:10.1038/s41419-020-2390-7.

19. Gupta, A.; Roy, S.; Lazar, A.J.; Wang, W.L.; McAuliffe, J.C.; Reynoso, D.; McMahon, J.; Taguchi, T.; Floris, G.; Debiec-Rychter, M.; et al. Autophagy inhibition and antimalarials promote cell death in gastrointestinal stromal tumor (GIST). Proc. Natl. Acad. Sci. USA 2010, 107, 14333–1438, doi:10.1073/pnas.1000248107.

20. Hsueh, Y.S.; Chang, H.H.; Chiang, N.J.; Yen, C.C.; Li, C.F.; Chen, L.T. MTOR inhibition enhances NVP-AUY922-induced autophagy-mediated KIT degradation and cytotoxicity in imatinib-resistant gastrointestinal stromal tumors. Oncotarget 2014, 5, 11723–11736, doi:10.18632/oncotarget.2607.

21. Hershey, J.W.; Sonenberg, N.; Mathews, M.B. Principles of translational control: An overview. Cold Spring Harb. Perspect. Biol. 2012, 4, a011528, doi:10.1101/cshperspect.a011528.

22. Delaidelli, A.; Jan, A.; Herms, J.; Sorensen, P.H. Translational control in brain pathologies: Biological significance and therapeutic opportunities. Acta Neuropathol. 2019, 137, 535–555, doi:10.1007/s00401-019-01971-8.

23. Jackson, R.J.; Hellen, C.U.; Pestova, T.V. The mechanism of eukaryotic translation initiation and principles of its regulation. Nat. Rev. Mol. Cell Biol. 2010, 11, 113–127, doi:10.1038/nrm2838. PMID: 20094052.

24. Sharma, D.K.; Bressler, K.; Patel, H.; Balasingam, N.; Thakor, N. Role of Eukaryotic Initiation Factors during Cellular Stress and Cancer Progression. J. Nucleic Acids 2016, 2016, doi:10.1155/2016/8235121.

25. Gingras, A.C.; Raught, B.; Sonenberg, N. eIF4 initiation factors: Effectors of mRNA recruitment to ribosomes and regulators of translation. Annu. Rev. Biochem. 1999, 68, 913–963, doi:10.1146/annurev.biochem.68.1.913.

26. Seeboeck, R.; Sarne, V.; Haybaeck, J. Current Coverage of the mTOR Pathway by Next-Generation Sequencing Oncology Panels. Int. J. Mol. Sci. 2019, 20, 690, doi:10.3390/ijms20030690.

27. Masvidal, L.; Hulea, L.; Furic, L.; Topisirovic, I.; Larsson, O. mTOR-sensitive translation: Cleared fog reveals more trees. RNA Biol. 2017, 14, 1299–1305, doi:10.1080/15476286.2017.1290041.

28. Naineni, S.K.; Itoua Maïga, R.; Cencic, R.; Putnam, A.A.; Amador, L.A.; Rodriguez, A.D.; Jankowsky, E.; Pelletier, J. A comparative study of small molecules targeting eIF4A. RNA 2020, 26, 541–549, doi:10.1261/rna.072884.119.

29. Chen, K.; Yang, J.; Li, J.; Wang, X.; Chen, Y.; Huang, S.; Chen, J.L. eIF4B is a convergent target and critical effector of oncogenic Pim and PI3K/Akt/mTOR signaling pathways in Abl transformants. Oncotarget 2016, 7, 10073–10089, doi:10.18632/oncotarget.7164.

30. Cencic, R.; Robert, F.; Galicia-Vázquez, G.; Malina, A.; Ravindar, K.; Somaiah, R.; Pierre, P.; Tanaka, J.; Deslongchamps, P.; Pelletier, J. Modifying chemotherapy response by targeted inhibition of eukaryotic initiation factor 4A. Blood Cancer J. 2013, 3, e128, doi:10.1038/bcj.2013.25.

31. Golob-Schwarzl, N.; Krassnig, S.; Toeglhofer, A.M.; Park, Y.N.; Gogg-Kamerer, M.; Vierlinger, K.; Schröder, F.; Rhee, H.; Schicho, R.; Fickert, P.; et al. New liver cancer biomarkers: PI3K/AKT/mTOR pathway members and eukaryotic translation initiation factors. Eur. J. Cancer 2017, 83, 56–70, doi:10.1016/j.ejca.2017.06.003.

32. Tapia, O.; Riquelme, I.; Leal, P.; Sandoval, A.; Aedo, S.; Weber, H.; Letelier, P.; Bellolio, E.; Villaseca, M.; Garcia, P.; et al. The PI3K/AKT/mTOR pathway is activated in gastric cancer with potential prognostic and predictive significance. Virchows Arch. 2014, 465, 25–33, doi:10.1007/s00428-014-1588-4.

33. Wang, L.; Wen, X.; Luan, F.; Fu, T.; Gao, C.; Du, H.; Guo, T.; Han, J.; Huang, F.L.; Cheng, X.; et al. EIF3B is associated with poor outcomes in gastric cancer patients and promotes cancer progression via the PI3K/AKT/mTOR signaling pathway. Cancer Manag. Res. 2019, 11, 7877–7891, doi:10.2147/CMAR.S207834.

34. Golob-Schwarzl, N.; Schweiger, C.; Koller, C.; Krassnig, S.; Gogg-Kamerer, M.; Gantenbein, N.; Toeglhofer, A.M.; Wodlej, C.; Bergler, H.; Pertschy, B.; et al. Separation of low and high grade colon and rectum carcinoma by eukaryotic

translation initiation factors 1, 5 and 6. Oncotarget 2017, 8, 101224–101243, doi:10.18632/oncotarget.20642.

35. Fresno Vara, J.A.; Casado, E.; de Castro, J.; Cejas, P.; Belda-Iniesta, C.; González-Barón, M. PI3K/Akt signalling pathway and cancer. Cancer Treat Rev. 2004, 30, 193–204, doi:10.1016/j.ctrv.2003.07.007.
36. Blanke, C.D.; Demetri, G.D.; von Mehren, M.; Heinrich, M.C.; Eisenberg, B.; Fletcher, J.A.; Corless, C.L.; Fletcher, C.D.; Roberts, P.J.; Heinz, D.; et al. Long-term results from a randomized phase II trial of standard- versus higher-dose imatinib mesylate for patients with unresectable or metastatic gastrointestinal stromal tumors expressing KIT. J. Clin. Oncol. 2008, 26, 620–625, doi:10.1200/JCO.2007.13.4403.
37. Yan, L.; Zou, L.; Zhao, W.; Wang, Y.; Liu, B.; Yao, H.; Yu, H. Clinicopathological significance of c-KIT mutation in gastrointestinal stromal tumors: A systematic review and meta-analysis. Sci. Rep. 2015, 5, 13718, doi:10.1038/srep13718.
38. Casali, P.G.; Zalcberg, J.; Le Cesne, A.; Reichardt, P.; Blay, J.Y.; Lindner, L.H.; Judson, I.R.; Schöffski, P.; Leyvraz, S.; Italiano, A.; et al. European Organisation for Research and Treatment of Cancer Soft Tissue and Bone Sarcoma Group, Italian Sarcoma Group, and Australasian Gastrointestinal Trials Group. Ten-Year Progression-Free and Overall Survival in Patients with Unresectable or Metastatic GI Stromal Tumors: Long-Term Analysis of the European Organisation for Research and Treatment of Cancer, Italian Sarcoma Group, and Australasian Gastrointestinal Trials Group Intergroup Phase III Randomized Trial on Imatinib at Two Dose Levels. J. Clin. Oncol. 2017, 35, 1713–1720, doi:10.1200/JCO.2016.71.0228.
39. Miettinen, M.; Lasota, J. Gastrointestinal stromal tumors: review on morphology, molecular pathology, prognosis, and differential diagnosis. Arch Pathol Lab Med. 2006, 130, 1466–1478, doi: 10.1043/1543-2165(2006)130[1466:GSTROM]2.0.CO;2.
40. Napolitano, A.; Vincenzi, B. Secondary KIT mutations: The GIST of drug resistance and sensitivity. Br. J. Cancer 2019, 120, 577–578, doi:10.1038/s41416-019-0388-7.
41. Demetri, G.D.; Reichardt, P.; Kang, Y.K.; Blay, J.Y.; Rutkowski, P.; Gelderblom, H.; Hohenberger, P.; Leahy, M.; von Mehren, M.; Joensuu, H.; et al. Efficacy and safety of regorafenib for advanced gastrointestinal stromal tumours after failure of imatinib and sunitinib (GRID): An international, multicentre, randomised, placebo-controlled, phase 3 trial. Lancet 2013, 381, 295–302, doi:10.1016/S0140-6736(12)61857-1.
42. Demetri, G.D.; Heinrich, M.C.; Fletcher, J.A.; Fletcher, C.D.; Van den Abbeele, A.D.; Corless, C.L.; Antonescu, C.R.; George, S.; Morgan, J.A.; Chen, M.H.; et al. Molecular target modulation, imaging, and clinical evaluation of gastrointestinal stromal tumor patients treated with sunitinib malate after imatinib failure. Clin. Cancer Res. 2009, 15, 5902–5909, doi:10.1158/1078-0432.CCR-09-0482.
43. Wilhelm, S.M.; Dumas, J.; Adnane, L.; Lynch, M.; Carter, C.A.; Schütz, G.; Thierauch, K.H.; Zopf, D. Regorafenib (BAY 73-4506): A new oral multikinase inhibitor of angiogenic, stromal and oncogenic receptor tyrosine kinases with potent preclinical antitumor activity. Int. J. Cancer 2011, 129, 245–255, doi:10.1002/ijc.25864.
44. Serrano, C.; Mariño-Enríquez, A.; Tao, D.L.; Ketzer, J.; Eilers, G.; Zhu, M.; Yu, C.; Mannan, A.M.; Rubin, B.P.; Demetri, G.D.; et al. Complementary activity of tyrosine kinase inhibitors against secondary kit mutations in imatinib-resistant gastrointestinal stromal tumours. Br. J. Cancer 2019, 120, 612–620, doi:10.1038/s41416-019-0389-6.
45. Patel, S. Exploring novel therapeutic targets in GIST: Focus on the PI3K/Akt/mTOR pathway. Curr. Oncol. Rep. 2013, 15, 386–395, doi:10.1007/s11912-013-0316-6.
46. Kasireddy, V.; von Mehren, M. Emerging drugs for the treatment of gastrointestinal stromal tumour. Expert Opin. Emerg. Drugs 2017, 22, 317–329, doi:10.1080/14728214.2017.1411479.
47. Kalfusová, A.; Kodet, R. Molecular mechanisms of primary and secondary resistance, molecular-genetic features and characteristics of KIT/PDGFRA non-mutated GISTs. Ceskoslovenska Patol. 2017, 53, 167–173.
48. Quan, P.; Moinfar, F.; Kufferath, I.; Absenger, M.; Kueznik, T.; Denk, H.; Zatloukal, K.; Haybaeck, J. Effects of targeting endometrial stromal sarcoma cells via histone deacetylase and PI3K/AKT/mTOR signaling. Anticancer Res. 2014, 34, 2883–2897.