bjh review

Current and emerging therapeutic approaches for T-cell acute lymphoblastic leukaemia

Rachael Pocock,[1] Nadine Farah,[1] Simon E. Richardson[2,3] and Marc R. Mansour[1]

[1]Department of Haematology, UCL Cancer Institute, University College London, London, [2]Wellcome-MRC Cambridge Stem Cell Institute, University of Cambridge, and [3]Department of Haematology, University of Cambridge, Jeffrey Cheah Biomedical Centre, Cambridge, UK

Summary

T-cell ALL (T-ALL) is an aggressive malignancy of T-cell progenitors. Although survival outcomes in T-ALL have greatly improved over the past 50 years, relapsed and refractory cases remain extremely challenging to treat and those who cannot tolerate intensive treatment continue to have poor outcomes. Furthermore, T-ALL has proven a more challenging immunotherapeutic target than B-ALL. In this review we explore our expanding knowledge of the basic biology of T-ALL and how this is paving the way for repurposing established treatments and the development of novel therapeutic approaches.

Keywords: leukaemia, novel treatments.

The need for new therapies in T-ALL was once driven by the inferior survival outcomes seen in T-ALL compared to B-ALL. Improvements in chemotherapy usage with treatment intensification and minimal residual disease (MRD) monitoring have made a major impact on this disparity.[1] The introduction of treatment escalation based on MRD has meant that despite a three-fold higher rate of positive MRD at end of induction (EOI) in T-ALL *versus* B-ALL,[2] both subtypes now have equivalent survival outcomes in children.[3] This is particularly relevant to early T-cell precursor T-ALL (ETP-ALL), historically associated with a very high-risk of treatment failure,[4] but now with excellent outcomes on MRD risk-directed protocols.[5-7] In adults, survival in T-ALL now surpasses B-ALL on some protocols.[8] Focus has also been directed to appropriate de-escalation of treatment in those with low risk MRD.[9]

Long term survival outcomes approach 50% in adults able to tolerate intensive treatment and exceed 90% in childhood ALL,[9-12] a remarkable prognosis that may reflect the superior tolerance of children to chemotherapy and a difference in the genetics of childhood leukaemia.[13,14] The vast majority of children that remain in remission 2 years from diagnosis will be cured, with rare cases of late relapse (>5 years) likely representing a clonally unrelated secondary T-ALL.[15] However relapsed T-ALL is highly aggressive and often resistant to glucocorticoids and chemotherapy, with survival of around 50% in children and less in adults, with the worst outcome in those with the shortest duration of remission.[16-19] For those adults who relapse, allogeneic transplant offers the best chance of cure, with recently reported survival outcomes of 40%.[19] Despite modern treatment protocols, durable responses for adults unable to proceed to transplant are unlikely, with a median survival of only 8 months.[19] In children, allogeneic bone marrow transplantation is generally reserved for those with high-risk relapsed disease and remains one of the few curative options for these patients.[20]

Genetic markers can identify good prognostic subgroups with potential for treatment de-escalation. Patients with both *NOTCH1* and *FBXW7* mutations, or two *NOTCH1* mutations, have been shown to have an excellent outcome, with 100% 5 years survival in this patient subgroup treated on the paediatric UKALL2003 trial,[21] and improved survival seen in adults.[22,23] However not all findings are as clear cut, for example PTEN aberrations have added additional prognostic value on some trials,[24-26] but not others.[27] Given the rarity of the disease, most studies are underpowered to detect small but meaningful differences in outcome among genetic subgroups. As an example, outcome analyses based solely on NOTCH pathway mutations alone are confounded by the high frequency of these mutations in TLX+ cases (approx. 90%) and relative rarity in ETP-ALL (approx. 20%).[28]

The poor outcomes seen in relapsed/refractory (R/R) T-ALL highlight a pressing need for novel treatments. Improved understanding of the genetics of both normal and aberrant T-cell differentiation is offering new therapeutic avenues. T-ALL is a genetically heterogeneous disease, which can be sub-classified based on first-hit class-defining lesions that commonly affect master regulatory transcription factors

Correspondence: Marc R. Mansour. Paul O'Gorman Building, UCL Cancer Institute, 72 Huntley Street, London. WC1E 6DD, UK.
Phone number: +44 2076796231.
E-mail: m.mansour@ucl.ac.uk

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.
doi: 10.1111/bjh.17310
This is an open access article under the terms of the Creative Commons Attribution License, which permits use, distribution and reproduction in any medium, provided the original work is properly cited.

(e.g. TAL1, LMO1/2, TLX1/3, HOXA). Transcription factors have proven extremely difficult to target pharmacologically, thus the focus of drug development has been on second hit mutations in key signalling pathways. Herein we review some current areas of active translational research in this field.

Nelarabine

Nelarabine is the soluble prodrug of ara-G, which is selectively cytotoxic to T leukaemic cells, likely due to their low endogenous SAMHD1 levels.[29] Initial early phase trials of nelarabine showed its efficacy as a single agent in paediatric relapsed/refractory T-ALL, with neurotoxicity the most common dose-limiting toxicity.[30,31] In adults, an alternate day dosing schedule limited neurotoxicity whilst maintaining an over all response rate (ORR) of 31%.[32] Subsequent data showed the efficacy of Nelarabine in combination with chemotherapy, with NEC (Nelarabine, Etoposide and Cyclophosphamide), with an impressive complete remission (CR) of 71% in relapsed patients.[33]

More recent data supporting the upfront use of Nelarabine in children and young adults has led some to consider it as standard of care in paediatric T-ALL.[34,35] Data for its upfront use in adults is awaited from the UKALL14 trial.

NOTCH inhibitors

NOTCH receptors are part of a conserved protein family that can act both as oncogenes or tumour suppressors, depending on the cellular context.[36] NOTCH1 is important for thymocyte development, committing common lymphoid progenitors to a T-cell fate. Activating mutations of *NOTCH1* have been found in almost two-thirds of paediatric and adult T-ALL cases.[37,38]

As one of the most frequently mutated genes in T-ALL, *NOTCH1* has generated considerable interest as a therapeutic target. Gamma secretase inhibitors (GSI), originally developed for Alzheimer's disease, act by preventing the cleavage and activation of the intracellular NOTCH1 fragment (Fig 1A). Their early promise has since been hampered by marked gastrointestinal (GI) toxicity.[39] NOTCH1 is an important regulator of intestinal goblet cells and NOTCH1 inhibition by GSIs causes goblet cell accumulation via upregulation of the transcription factor *KLF4*, resulting in significant diarrhoea (Fig 1B).[40] However, the use of a pulsed treatment schedule[41] and the addition of glucocorticoids have reduced this toxicity,[42] with one reported case of a CR in a patient treated with a GSI and dexamethasone.[43] Not only do glucocorticoids improve the side effect profile of GSIs, but also the two treatments work synergistically to induce apoptosis of T-ALL cells, possibly due to increased expression of the glucocorticoid receptor NR3C1 in the presence of the combination.[42] However, a recently reported phase 1 trial using a novel inhibitor of the NOTCH ICD in combination with dexamethasone continued to show dose limiting GI toxicity and limited clinical efficacy.[44] A more recent approach targeting PSEN1 aims to reduce the systemic toxicity associated with GSIs.[45] This subunit of the gamma-secretase complex is more highly expressed in leukaemic cells than normal developing T cells and its inhibition has been well tolerated in animal studies.

The NOTCH transcriptional co-activator MAML1 is also a potential target. Stapled α-helical peptides derived from MAML1 (SAHM) compete with MAML1 and inhibit NOTCH1-driven transcription. Mice treated with SAHM show a decrease in leukaemic cell burden and corresponding reduction in NOTCH1 target gene expression, including *MYC*.[46] Additionally their side effect profile seems tolerable in animal models, without the GI toxicity associated with GSIs.

Another method of modulating NOTCH involves the use of the proteasome inhibitor bortezomib, a standard of care treatment in myeloma. Bortezomib has activity in relapsed/refractory T-ALL, potentially by inhibiting transcriptional expression of NOTCH1.[47] In a small cohort of children with relapsed/refractory ALL, bortezomib appeared to have particular efficacy in T-ALL with complete response rates of over 70% when used in conjunction with chemotherapy.[48]

There are other considerations that have clinical relevance for NOTCH inhibition. Firstly, *NOTCH1* mutations are often late secondary subclonal events,[14,49] meaning NOTCH inhibitors are highly likely to select for *NOTCH1* wild-type cells. Secondly, resistance mechanisms can emerge whereby cells are able to maintain MYC levels in the absence of *NOTCH* signalling, for example through loss of FBXW7 or use of an alternative *MYC* enhancer.[50,51] In the former study, enhancer usage switches from the *NOTCH-MYC* enhancer to a *BRD4* regulated *MYC* enhancer, providing a rationale for combining NOTCH and BRD4 inhibitors.[50]

PI3K inhibitors

The PI3K-mTOR pathway plays a key role in both normal T cell and malignant cell development. Phosphoinositide 3 kinases (PI3K) are a family of lipid kinases that act as second messengers and are broadly divided into three classes, which share a common core PI3K motif (Fig 2). Almost half of T-ALL cases have aberrant PI3K activation occurring through deletion or mutation or *PTEN*, activating mutations of *PIK3R1* (typically N564D), *PIK3CD* (typically E1021K), or loss of function mutations of *USP7*.[28] These mutations are particularly enriched in the TAL1 subgroup,[28] with these two oncogenic pathways shown to synergise in mouse models, possibly related to the ability of AKT to phosphorylate and modulate TAL1 activity.[52] This raises the possibility that the TAL1 subgroup could be particularly susceptible to PI3K pathway modulation, although this awaits further study.

Mutations in the PI3K pathway also correlate with response to chemotherapy; homozygous deletions of *PTEN* appear to confer a higher risk of early treatment failure[53] and mutations in PTEN are associated with primary glucocorticoid

 © 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

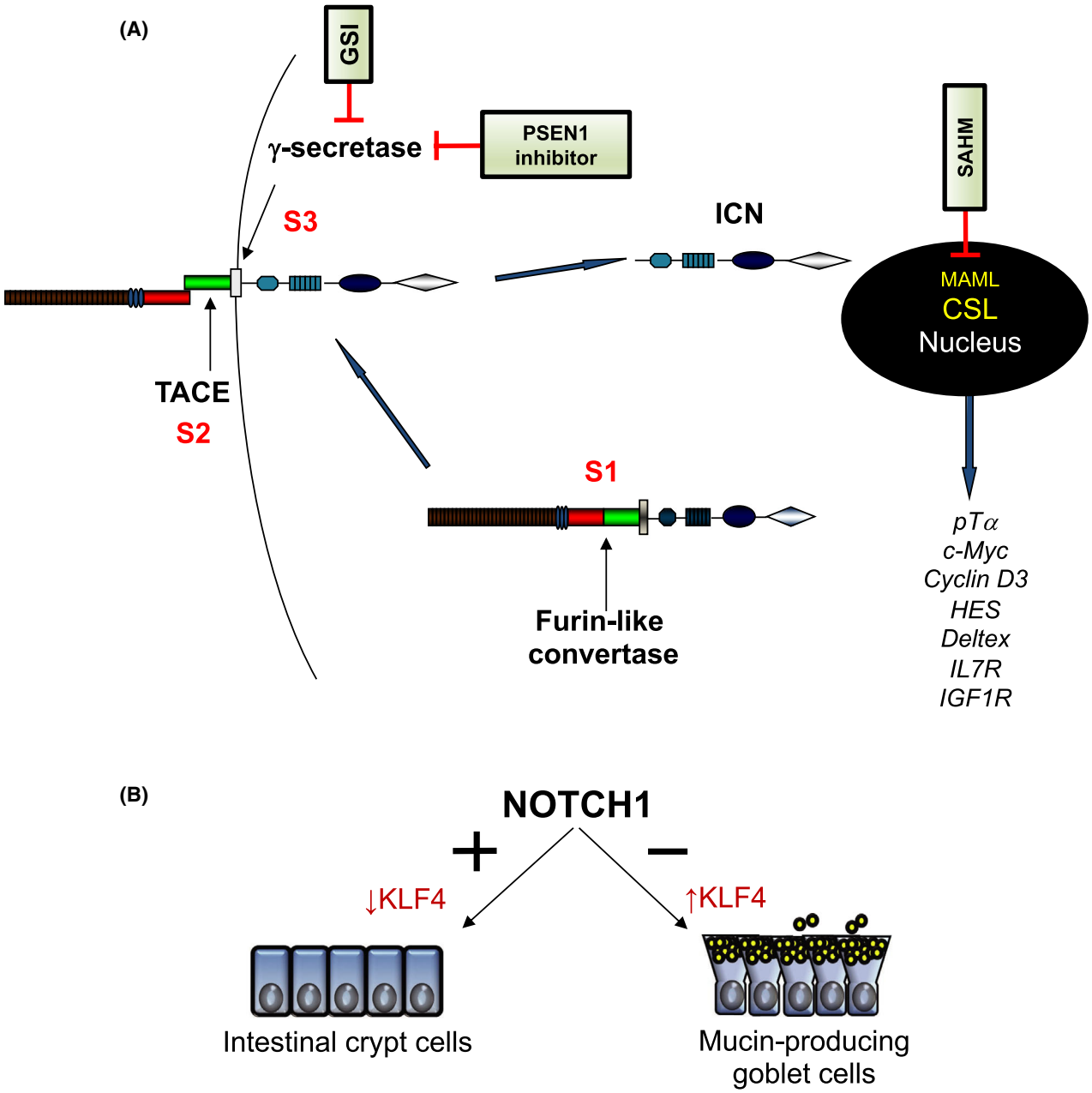

Fig 1. (A) NOTCH is activated by three cleavage steps. In the golgi, NOTCH1 is first cleaved in the Heterodimerization (HD) domain by a furin-like convertase (S1 cleavage) and held together by a non-covalent bond. On activation by ligand, the Lin-12/NOTCH repeats (LNR) domain is pulled from the Heterodimerization (HD), exposing the S2 cleavage site to proteolytic cleavage by TNFα-converting enzyme (TACE). This triggers S3 cleavage by the γ-secretase complex in the transmembrane domain releasing ICN to translocate to the nucleus to bind CSL and the transcriptional coactivator MAML activating a multitude of target genes, some of which are shown. (B) NOTCH1 determines intestinal cell fate. NOTCH1 inhibition results in accumulation of mucin-producing goblet cells through a KLF4 dependent pathway, resulting in severe diarrhoea.[40] [Colour figure can be viewed at wileyonlinelibrary.com]

resistance.[54] This is likely driven by AKT1-mediated phosphorylation of the glucocorticoid receptor, leading to impaired nuclear localisation; T-ALL mouse models treated with glucocorticoids and an AKT inhibitor, showed an augmented anti-leukaemic response, compared to mice treated with either agent alone.[55] Another synergistic approach looked to target both PI3K and NOTCH when it was found that T-ALL cells evade cell death when treated with PI3K/mTOR inhibition by upregulating NOTCH target genes such as *MYC*.[56]

The mTOR inhibitor everolimus acts downstream of AKT and in addition to chemotherapy, gave a 50% response rate in a small cohort of heavily pre-treated R/R T-ALL patients[57] it is currently being evaluated in a phase I trial with NEC (NCT03228104).

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

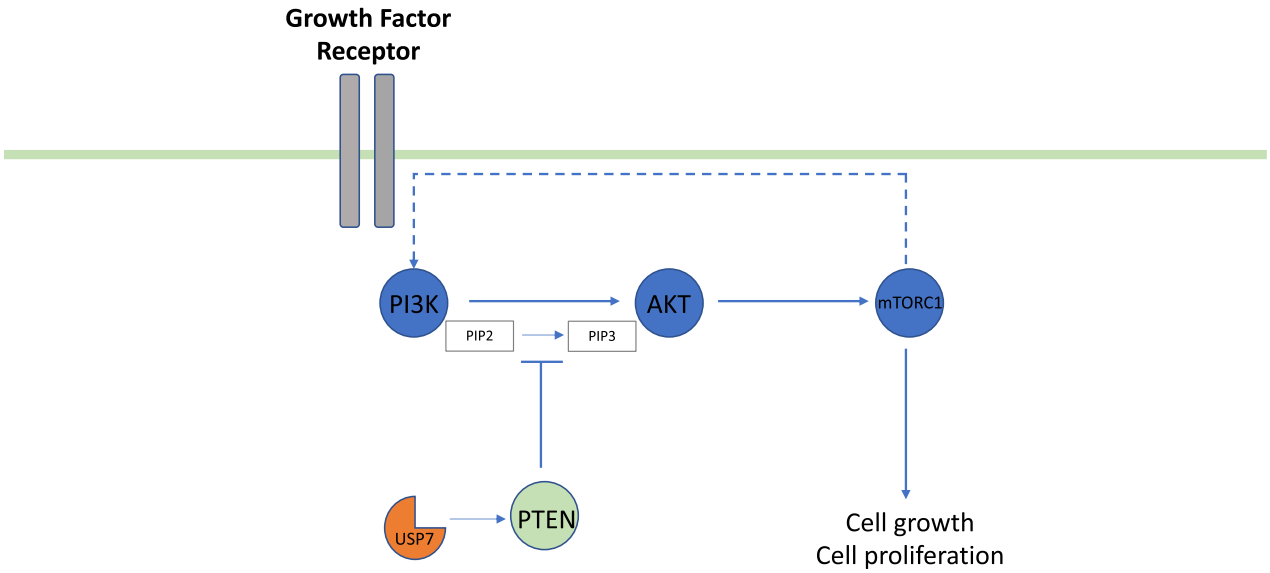

Fig 2. The PI3K signalling pathway: PI3K is activated by several growth factor receptor tyrosine kinases or G protein-coupled receptors. Once activated PI3K catalyses the phosphorylation of phosphatidylinositol 4,5-bisphosphate (PIP_2) to generate phosphatidylinositol 3,4,5-trisphosphate (PIP3). This activates the serine/threonine kinase AKT, which then drives mTOR to promote cell proliferation and survival. Dashed line indicates the negative feedback loop involving mTORC1 and PI3K. The ubiquitin specific peptidase USP7 acts to stabilise PTEN, a negative regulator of the PI3K pathway.

During normal lymphopoiesis, lymphocytes with hyperactive responses undergo negative selection through over-activation of PI3K signalling.[58,59] Thus, one concept gaining traction is that TCR stimulation, such as through an activating CD3-targeting monoclonal antibody, rather than inhibition of PI3K signalling may be an exploitable route towards initiating apoptosis in ALL cells.[60] A potential concern of this approach would be cytokine release syndrome, as occurred previously in solid organ transplant studies using OKT3.[61]

IL7R-JAK-STAT inhibition

The IL7R-JAK-STAT pathway is responsible for transducing cytokine signalling in the thymus and is required for normal T-cell development. This pathway is frequently aberrantly activated in the TLX1/3+ T-ALL subgroup, but very rarely in TAL1+ T-ALL. Activating mutations occur at multiple levels in the pathway, with recurrent mutations described in *STAT5B*, *IL7R*, *JAK1* and *JAK3*.[62-66] Interestingly, *JAK1* mutations appear to be more common in adult than paediatric T-ALL, and have been associated with reduced overall survival and a high relapse rate.[67] Mice transplanted with progenitor cells harbouring the most commonly identified *JAK3* mutation (M511I) develop an immature T-ALL through activating JAK1.[68]

The majority of activating *IL7R* mutations described thus far in T-ALL involve the insertion of a cysteine in the transmembrane domain leading to disulphide bonding and ligand-independent IL7R homodimerization and JAK1 phosphorylation (Fig 3).[62,64,67] Inhibition of the IL7R-JAK-STAT pathway has shown efficacy using clinically available JAK inhibitors in preclinical models[69,70] and phase I/II clinical trials of Ruxolitinib are planned. The disulphide bond and homodimer can also be disrupted by the reducing agent N-acetylcysteine (NAC) at doses readily achievable in patients.[71] Given its affordability and widespread use in treating patients with paracetamol overdose, such an approach would be particularly attractive in healthcare systems where targeted agents are considered prohibitively expensive.

An additional aspect of JAK inhibition is the possibility of restoring glucocorticoid sensitivity.[72,73] This approach is particularly intriguing in ETP-ALL, since ETP-ALL tends to be less sensitive to steroids than other T-ALL subtypes and STAT5 is commonly activated even in the absence of clearly identified upstream mutations.[69,74]

The long-term efficacy of JAK inhibition in T-ALL is unclear. Treatment does not eradicate leukaemic cells *in vitro*, leading to rapid relapse upon drug withdrawal, and combination therapy is likely to be required.[72] Moreover, due to the specific site of JAK inhibitor binding, it is possible that T-ALL cells will acquire resistance mutations, for example in the ATP-binding pocket of the kinase domain, or with mutations activating other JAK family members.[75] Most clinical JAK inhibitors act to competitively inhibit the ATP-binding pocket of the active JAK2 (Type 1), however there is growing interest in Type 2 JAK inhibitors which bind the ATP pocket of the inactive JAK, as well as a less-conserved nearby allosteric pocket.[76] This approach may provide a route to avoid resistance. Another strategy is to target further downstream of the IL7R-JAK pathway, such as the kinase PIM1. PIM1 appears to be upregulated in response to chemotherapy and steroids in a ($CD127^+$)[77] subset of T-ALL

 © 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

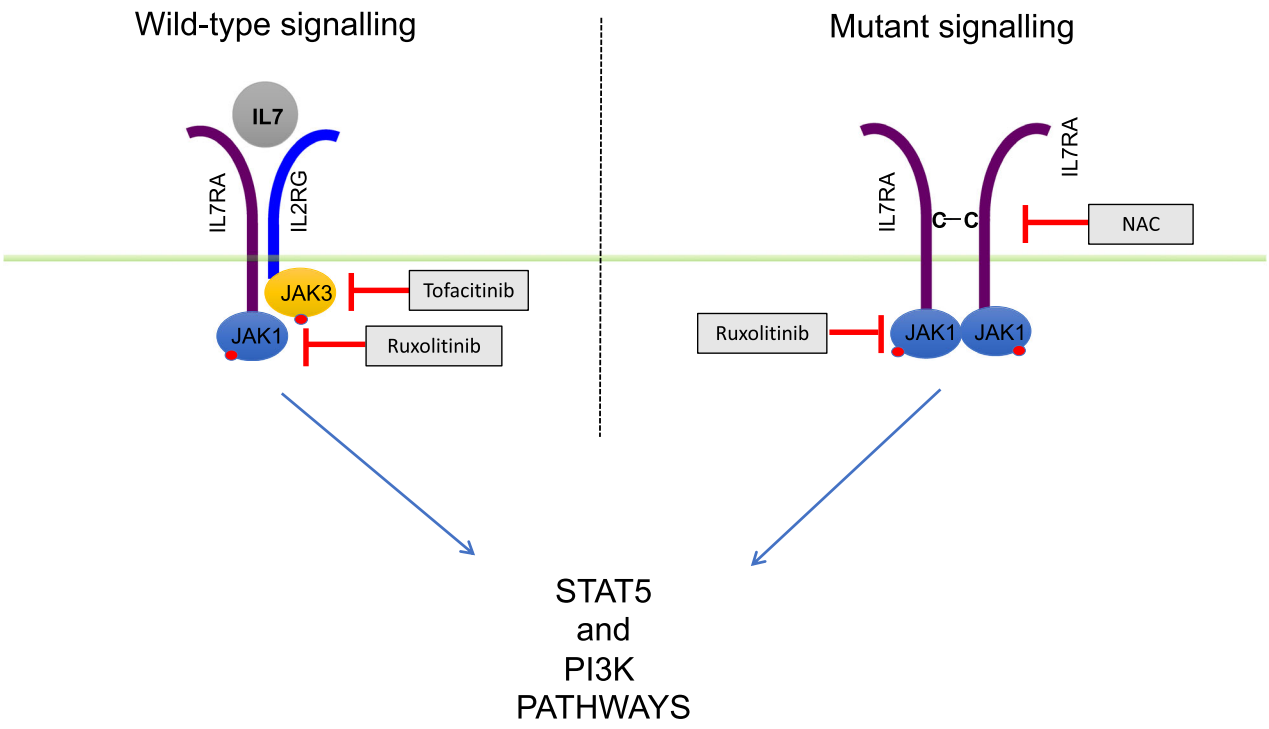

Fig 3. Schematic of anti-apoptotic dependencies in T-ALL according to level of differentiation arrest. ETP-ALL arrested early in T-cell development are highly dependent on BCL2, with increasing dependency on BCL-XL in T-ALLs that arrest Adapted from Chonghaile et al. (reference 79) in the cortical/post-cortical stages. This corresponds to vulnerability to Venetoclax and Navitoclax respectively. ETP – early T-cell progenitor; ISP immature single progenitor; EDP early double positive; DP double positive; SP single positive T-cell.

cells and PIM1 inhibition may be an alternative or adjunct to JAK inhibition. Such inhibitors are already in clinical trials for myelofibrosis.

Targeting anti-apoptotic machinery

B Cell lymphoma 2 (BCL2) is an anti-apoptotic protein that was first discovered from cloning the t(14;18) translocation in a case of B cell lymphoma. BCL2 forms part of a protein family that share the BCL2 homology (BH) domain. BH3 proteins, such as BAX and BAK, are pro-apoptotic proteins that are sequestered by BCL2, BCL-XL and MCL1.[78] More mature T-ALL cell lines have been shown to be dependent on BCL-XL, whereas ETP-ALL cells show a greater dependency on BCL2 (Fig 4).[79,80] Accordingly, ETP-ALL models have shown particular sensitivity to Venetoclax over Navitoclax, with reversal of this pattern in more mature T-ALL cells.[79] There is considerable clinical experience with Venetoclax in the treatment of CLL and more recently in AML, where the drug is generally well-tolerated. There is accumulating evidence for its use in T-ALL; a recent retrospective report of the use of Venetoclax with chemotherapy in R/R T-ALL showed that 6/13 patients achieved a morphological remission.[81] We suggest that Venetoclax should be a priority for incorporation in upcoming clinical trials, for instance in patients with high risk MRD at end of induction, or those with relapsed/refractory disease.

Navitoclax binds preferentially to BCL-XL, with less potent activity against BCL-2.[82] Its initial promise in CLL was limited by thrombocytopaenia,[83] as platelet survival requires normal function of BCL-XL. Despite this, it warrants testing in relapsed/refractory cortical and post-cortical T-ALL, where intrinsic mitochondrial chemoresistance is often the major barrier to achieving remission, although thrombocytopenia will need to be cautiously managed. Recent early phase trials of combination Venetoclax and Navitoclax are promising: In a small, heavily pre-treated patient cohort 50% of the T-ALL patients achieved CR/CRi (EHA 2020 S116). MCL1 is also an important target, since it has been associated with steroid resistance and poor outcome in T-ALL, directly upregulated by a recently discovered T-ALL oncogene called JDP2.[84] Direct MCL1 inhibitors such as S63845 have shown pre-clinical efficacy in T-ALL and are in early phase trials.[85] Alternative strategies that downregulate MCL1 expression, such as the CDK9 inhibitor AZD4573, also offer exciting new therapeutic opportunities across diverse haematological cancers.[86]

Several mechanisms of acquired resistance to Venetoclax have been described, including mutations of the drug binding site, deletions/mutations of BIM/BAX, and upregulation of BCL-XL and MCL1.[87] There is thus considerable interest in using Venetoclax in combination with Navitoclax, or MCL1 inhibitors, though how such combinations will be tolerated in terms of toxicity will need to be carefully assessed.

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

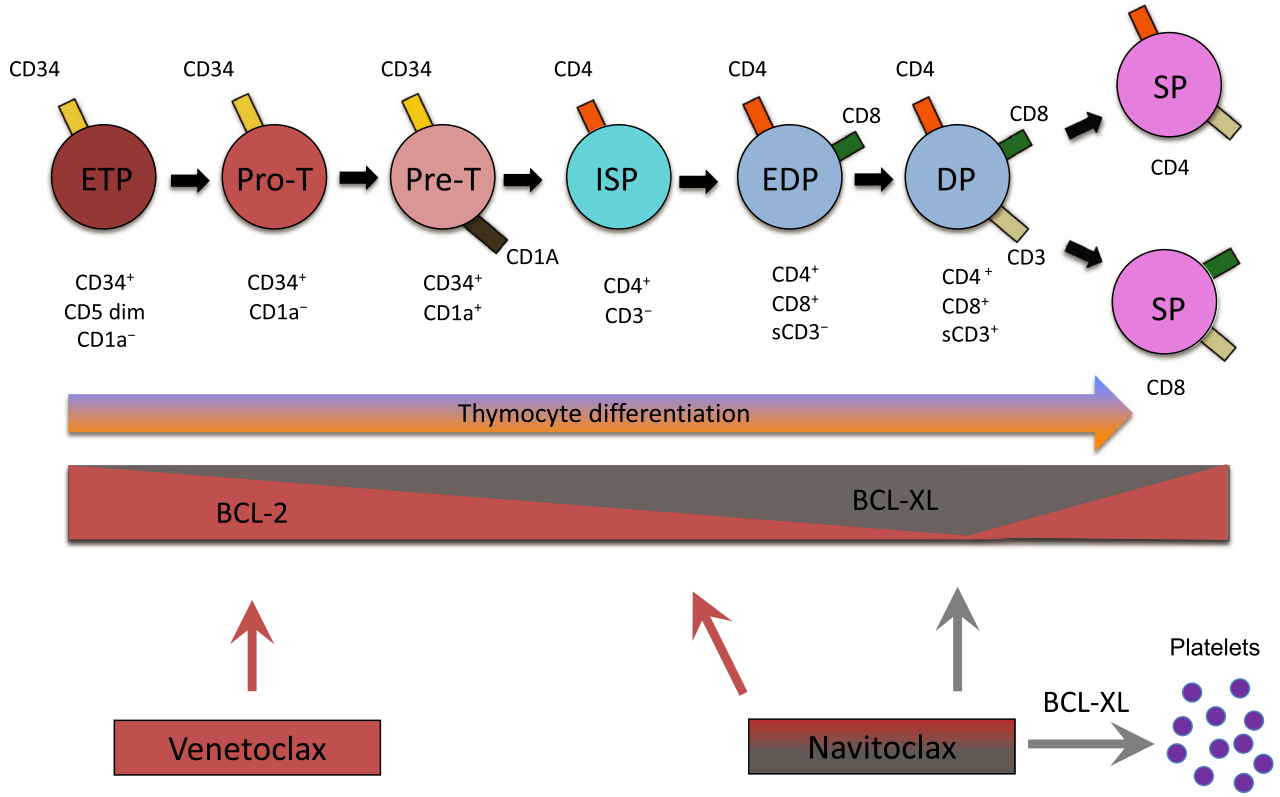

Fig 4. Schematic of anti-apoptotic dependencies in T-ALL according to level of differentiation arrest. ETP-ALL arrested early in T-cell development are highly dependent on BCL2, with increasing dependency on BCL-XL in T-ALLs that arrest Adapted from Chonghaile et. al. (reference 79) in the cortical/post-cortical stages. This corresponds to vulnerability to Venetoclax and Navitoclax respectively. ETP – early T-cell progenitor; ISP immature single progenitor; EDP early double positive; DP double positive; SP single positive T-cell.

Tyrosine kinase inhibitors

Aberrant tyrosine kinase activation in T-ALL occurs when chromosomal translocations involving the *ABL1* oncogene result in ligand independent activation of the *ABL1* kinase.[88] However, unlike chronic myeloid leukaemia (CML), the Philadelphia chromosome has only occasionally been reported in T-ALL. Instead the *ABL1* oncogene is fused with other partners, typically *NUP214* leading to constitutively active kinases.[89] These fusion products are still amenable to inhibition with tyrosine kinase inhibitors (TKIs), a class of drug that has transformed the outcomes of CML and Ph+ALL. *NUP214-ABL1* T-ALL cells, found in the TLX1/3 subgroups, respond to treatment with Imatinib, Nilotinib and Dasatinib.[90] These results have been recapitulated *in vivo* in a *NUP214-ABL1* T-ALL xenograft when imatinib treatment resulted in a reduction of leukaemic cell burden, where the addition of Venetoclax further improved the response.[91]

Whilst *ABL* translocations only account for approximately 5% of T-ALL cases, functional drug testing has unexpectedly revealed that up to 30% of T-ALL cases are susceptible to the ABL/SRC family kinase inhibitor Dasatinib.[92] No correlation was noted between responsiveness and established genetic lesions, including *ABL* translocations, leading the authors to propose that Dasatinib was targeting SRC, rather than ABL. This hypothesis was consistent with the lack of activity shown by other canonical ABL family kinase inhibitors, such as Imatinib, that do not affect SRC signalling. The *in vitro* activity of Dasatinib was confirmed by *in vivo* testing and supported by case reports of T-ALL responders to Dasatinib.[93] These findings are strengthened by those of another group who used *in silico* drug screening to identify up-regulation of the SRC family kinase *LCK* as a possible therapeutic target in T-ALL and demonstrated preclinical efficacy of Dasatinib.[94] Recently published data suggests synergy between Dasatinib and dexamethasone mediated via LCK.[95] Overall, it is likely that TKIs will have significant clinical activity in T-ALL, but their clinical use will remain limited until the identification of validated biomarkers.

Cyclin dependent kinase inhibitors

Cyclin dependent kinases (CDKs) are a large family of kinases with diverse roles, including acting as transcriptional co-factors and controlling cell cycle progression. Clinically meaningful inhibition of CDKs has proved technically challenging due to their integral role in normal cell survival and difficulties in targeting specific kinase isoforms. However, a

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

series of more specific small molecule inhibitors have recently emerged.

D cyclins are key cell cycle regulators that bind and activate CDK4 and 6, leading to activation of the E2F transcription factors that facilitate cell-cycle progression. Cyclin D3 is dysregulated in T-ALL and has been shown to be integral to NOTCH driven leukaemogenesis;[28] mice lacking cyclin D3 are resistant to NOTCH-driven transformation to T-ALL.[96] Furthermore, in a mouse model of T-ALL driven by activating mutations of *NOTCH1*, conditional ablation of cyclin D3 resulted in marked disease regression, findings that were phenocopied by exposure to the Cyclin D-CDK4/6 kinase inhibitor.[97] Efficacy of CDK4/6 inhibition has also been demonstrated in *NOTCH1* wildtype T-ALL cell lines where an *in vivo* model showed synergism with steroids and mTOR inhibitors.[98] Several CDK4/6 inhibitors are now in clinical trials and the challenge remains to identify targetable interdependencies between specific CDK isoforms and different mutational drivers in order to synergise their anti-leukaemic properties. In this regard, it will be important to assess whether the recently discovered recurrent mutations of CCND3 are a biomarker for sensitivity to CDK4/6 inhibition.[28]

CDK7 is a key constituent of the cyclin-activating kinase (CAK) complex, which acts to modulate the cell cycle by interacting with the general transcription factor, TFIIH. The CAK complex promotes transcription by activating RNA polymerase II (RNAPII) via CDK7-dependent phosphorylation. The novel agent THZ1 can specifically and irreversibly inhibit CDK7 by covalently binding to an amino acid located outside its kinase domain,[99] resulting in cell death in T-ALL cell lines (Fig 5). Interestingly, the activity of this agent may be associated with its preferential inhibition of super-enhancer driven oncogenes, such as the non-coding mutations that drive *TAL1*, providing a potential *in vivo* therapeutic window.[99,100]

CDK9 regulates transcriptional elongation and is of interest as an anti-cancer target in a range of malignancies, particularly those dependent on MCL1 (Fig 5). Targeted inhibition had initially been challenging due to homology of its ATP-binding site with other CDKs, until the development of the specific inhibitor AZD4573.[86] An alternative PROteolysis Targeting Chimeras (PROTAC) approach was recently developed where THAL-SNS-032 targeted CDK9 protein for proteosomal degradation, inducing apoptosis in T-ALL cell lines *in vitro*.[101]

Drugs in development

In this section we briefly explore further potential therapies that may emerge from encouraging preclinical data.

The Hedgehog pathway is an evolutionarily conserved signalling cascade, however its aberrant activation also drives tumour growth and chemoresistance. This pathway is activated in up to a fifth of T-ALL and is associated with induction of chemotherapy resistance.[102] Inhibition of GLI1, a Hedgehog pathway transcription factor, resulted in improved survival in T-ALL PDX models and could offer a novel target for high-risk T-ALL.[103]

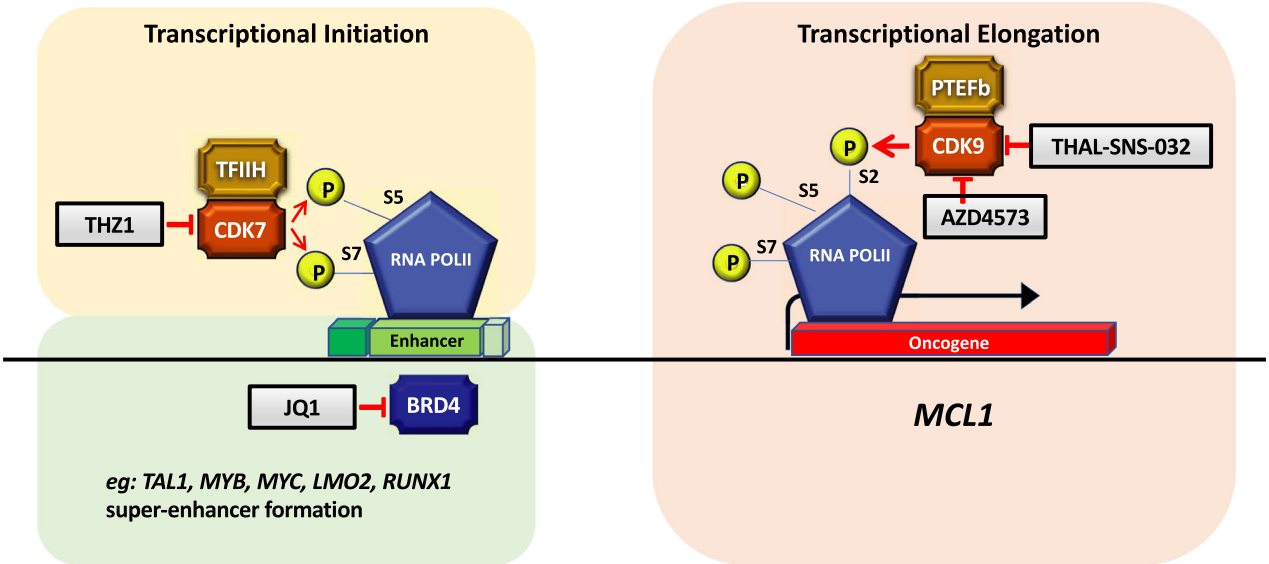

Fig 5. CDK7 and CDK9 regulate oncogenes and are therapeutic targets in T-ALL. Green box: super-enhancers driving oncogenes can be destabilised by inhibition of chromatin interacting proteins such as BRD4. Yellow box: The initiation of RNA polymerase II (RNApolII) mediated oncogene transcription is facilitated by phosphorylation of the C terminal tail on serine 5 and 7 by CDK7 as part of the TFIIH complex, which occurs both at enhancers and gene start sites. CDK7 can be inhibited by small molecules such as THZ1. Red box: Transcriptional elongation by RNApolII is facilitated by phosphorylation of RNApolII on serine 2 by CDK9 as part of the PTEFb complex. CDK9 can be targeted for degradation by the novel agent THAL-SNS-032 or inhibited by CDK9 inhibitors such as AZD4573. CDK9 inhibition has been associated with marked downregulation of MCL1 expression.

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

Histone deacetylases (HDAC) play a role in the regulation of chromatin structure and the HDAC inhibitors (HDACi) Panobinostat, Vorinostat and Romidepsin are already in clinical use in myeloma and lymphoma. The broad-acting HDACi panobinostat shows particular efficacy *in vivo* T-ALL models with improved survival seen in combination with chemotherapy.[104] Interestingly, there is a suggestion that the efficacy of panobinostat over other HDACi relates to its epigenetic inhibition of the oncogene *MYC*.[105]

Exportin 1 (XPO1) (also known as CRM1) is a nuclear-cytoplasmic exporter protein involved in the transport of several proteins involved in cell cycle regulation. It is the only transporter of key tumour suppressors including TP53 and is upregulated in several malignancies, including T-ALL. Selinexor, a selective inhibitor of nuclear export (SINE) compound, is a small molecule XPO1 antagonist and is FDA-approved for myeloma. It has shown preclinical efficacy in T-ALL, although toxicity reported from clinical trials may prove problematic.[106]

Heat shock proteins act as molecular chaperones for a variety of proteins with key roles in oncogenesis, including the JAK pathway and another, closely related kinase pathway, TYK2. TYK2 acts to upregulate BCL2 via STAT1[107] and its inhibition by the drug Luminespib/AUY922 triggers apoptosis in T-ALL *in vitro*. It appears that this effect is mediated via a reduction in TYK2 and subsequent downregulation of BCL2.[108] Early clinical trials of Luminespib in a range of malignancies have been undertaken and this could yet be an additional therapy in T-ALL.

Approximately 75% of proteins currently have no targetable domain. PROteolysis Targeting Chimeras (PROTACs) circumvents this problem by degrading proteins instead of inhibiting them, thus broadening the number of potential targets. This approach may be particularly relevant to diseases such as T-ALL where the majority of driver translocations involve oncogenic transcription factors.

Another technique looks to change the method of drug delivery, using nanoparticles, enabling optimized drug dosing and potentially combining chemotherapy with a targeted ligand. This remains a broadly experimental area of leukaemic treatment, however the success of CPX-351 (a liposomal formulation of cytarabine and daunorubicin) in AML demonstrates the potential of this expanding therapeutic area.

Immunotherapy

Immunotherapies are a group of therapeutics that harness the immune system to specifically attack malignant cells. Broadly they can be divided into treatments that: (i) amplify a natural anti-tumour immune response (e.g. immune checkpoint blockade); or (ii) synthetic immunotherapies (e.g. monoclonal antibodies or chimeric antigen receptor T or NK cells). There have been limited studies of immune checkpoint blockade in ALL and the relatively low mutational burden seen, particularly in paediatric ALL may limit the expression of tumour specific neo-antigens, on which this strategy is thought to rely. By contrast, synthetic immunotherapies are having a major impact in the treatment of B-ALL with recent clinical advances including the use of Rituximab, Inotuzumab, Blinatumomab and durable responses to anti-CD19 chimeric antigen receptor (CAR)-T cell therapy.[109]

All of these successful synthetic immunotherapies rely on the presence of an antigen that is strongly expressed on all leukaemic blasts, but with limited expression on normal tissues. In the case of B cells, strong expression of specific B cell markers such as CD19, CD20 and CD22 have provided good immune targets. Furthermore, whilst normal B cells are also attacked, B cell aplasia is clinically manageable and therefore precision targeting of leukaemic B cells is not an absolute requirement. By contrast, immunotargeting T-ALL has a number of technical hurdles Fig 6. Firstly, antigen expression is variable at different stages of T cell differentiation, and thus there is not a single antigenic target that is likely to be applicable to all T cell malignancies. Secondly, immune destruction of normal T cells would result in a life-threatening immune deficiency disorder. Thirdly, harvesting of autologous T cells for CAR T cell generation risks contamination by T-ALL blasts. Lastly, CAR T cells expressing the same antigen they are targeting would lead to fratricide and T-cell exhaustion. There is therefore limited clinical experience of synthetic immunotherapies in T-ALL, but a number of promising approaches are emerging.

Monoclonal antibodies

One of the most promising candidates in the short term is the anti-CD30 drug immunoconjugate Brentuximab Vedotin, which is licensed for use in classical Hodgkin lymphoma and anaplastic large cell lymphoma. Preclinical studies have shown that CD30 is expressed in 13/34 of T-ALL cases tested by flow cytometry, but pre-clinical evidence for functional efficacy is lacking.[110] As some T-ALL cases, typically of the ETP subgroup, express CD33, there is also the potential to use Gemtuzumab Ozogamicin in such cases, but clinical data for this approach in T-ALL has not been reported. Another potential surface target in T-ALL is the activation marker CD38, which is highly expressed on T-ALL blasts and targetable by the monoclonal antibody daratumumab. Pre-clinical efficacy of Daratumumab in T-ALL has translated into clinical responses in four post-allogeneic relapsed/refractory cases who achieved MRD negativity, an encouraging set of results for such aggressive disease[111-113] and the Delphinus phase II clinical trial is currently recruiting in the UK (NCT03384654).

The early lymphoid cytokine receptor and T-ALL oncogene IL7R is a potentially attractive immunotherapy target. Recent work has preclinically explored a novel human monoclonal antibody targeting IL7R, showing evidence that binding inhibits signalling, functionally sensitises cells to glucocorticoids and elicits natural killer (NK) cell mediated cellular cytotoxicity *in vitro*, and potentially *in vivo*.

 © 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

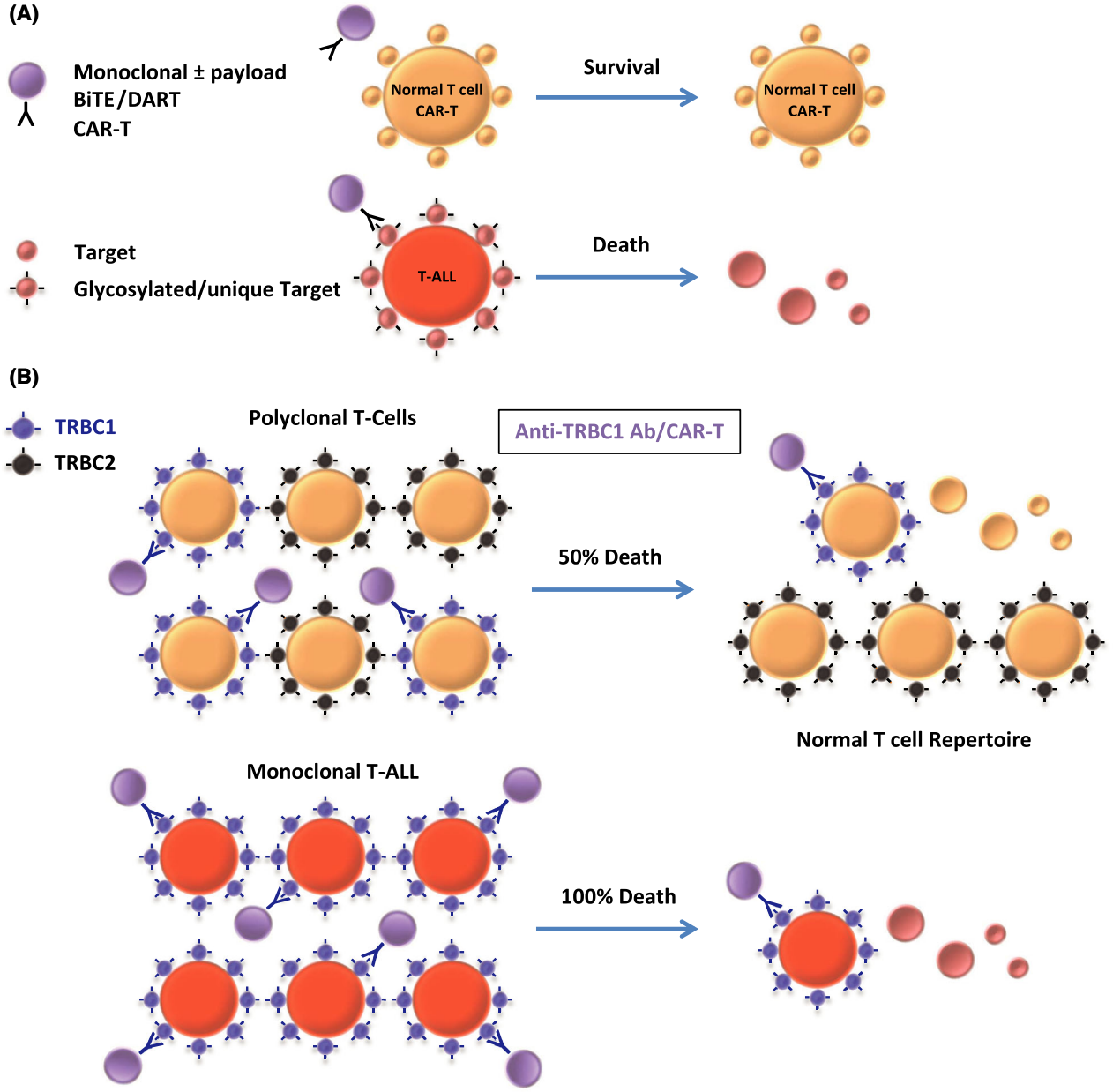

Fig 6. Potential immunotherapy approaches for T-ALL. (A) T-ALL specific neo-epitopes can be targeted by monoclonal antibodies (with or without conjugated cytotoxic payloads) or used to target cellular cytotoxicity via T cell engagers (e.g. bi-specific T cell engagers (BiTE) or dual affinity retargeting (DART)) or chimeric antigen T cells (CAR T); (B) targeting TRBC1 on TRBC1-expressing monoclonal T-ALL overcomes CAR T fratricide and T cell immunodeficiency by affording the retention of a polyclonal T cell repertoire of TRBC2-expressing normal and CAR T cells.

Furthermore, the antibody is internalised on binding, giving a potential utility in chemo-immunotherapy.[114] Whilst providing a platform for a number of promising approaches, the potential for on- and off-target toxicities remain to be seen.

Chimeric antigen receptor cells

A number of CAR T or NK cell approaches have shown benefit in pre-clinical studies, but there is no single antigenic target that is expressed in all T-ALL cases. An anti-CD4 CAR T cell has shown activity against a $CD4^+$ T-ALL cell line in a xenograft model and this approach circumvents CAR T cell fratricide by sparing cytotoxic $CD8^+$ effector cells.[115] However, only a minority of T-ALL cases express CD4 and prolonged CD4 aplasia is likely to lead to life-threatening infection.

An anti-CD3 CAR-NK cell line has shown activity against a T-ALL cell line in a xenograft model.[116] The use of a CD3

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

Table I. International clinical trials.

Trial number	Countries recruiting	Disease status	Phase	Treatment	Age	Current status	Est. closing date
Nelarabine							
NCT03328104	USA	Relapsed/refractory	I	Everolimus + nelarabine (+etoposide and cyclophosphamide)	2–29 years	Recruiting	Oct 2020
NCT00501826	USA	Untreated	II	Nelarabine + Hyper CVAD + Pegaspargase + venetoclax	Child/Adult	Recruiting	Oct 2020
NCT02763384	USA	Relapsed/refractory	II	Nelarabine + BL8040 (CXCR4 antagonist)	>18 years	Recruiting	May 2021
NCT02619630	France	New diagnosis	II	Nelarabine (+cyclophosphamide + etoposide)	18–59yrs	Recruiting	Dec 2020
NCT02881086	Germany	New diagnosis	III	Nelarabine (+PEG asparaginase/ dex/cyclophosphamide/ methotrexate/cytarabine/vindesine/ adriamycin/prednisolone)	18–55	Recruiting	July 2021
Tyrosine kinase inhibitors							
NCT01620216	USA	Relapsed/refractory inc AML	II	(Dasatinib v ponatinib v sorafenib v nilotinib v sunitinib)	>18 years	Unknown	March 2019
Janus kinase inhibitors							
NCT03613428	China	Relapsed/refractory ETP-ALL	I/II	Ruxolitinib (+vincristine, prednisolone, L-asparaginase)	13–75 years	Not yet recruiting	Dec 2020
CDK6 inhibitor							
NCT03515200	USA	Relapsed/refractory	I	Palbociclib (CDK4/6 inhibitor)+ dexamethasone, bortezemib, dasatinib, doxorubicin	<22 years	Terminated	July 2020
NCT03263637	Germany/Netherlands/UK	Relapsed/refractory	I	AZD4573 (CDK9 inhibitor)	18–130 years	Active, not recruiting	Dec 2021
BCL inhibitor							
NCT03181126	USA/Australia	Relapsed/refractory	I	Navitoclax + venetoclax + chemo	4–16 years	Active, not recruiting	Nov 2020
NCT03236857	USA/Australia/Canada/ France/Germany/ Netherlands/Switzerland/UK	Relapsed/refractory	I	Venetoclax (+/-chemo)	<25 years	Recruiting	April 2022
NCT03808610	USA	Relapsed/refractory	II	Venetoclax + low dose chemotherapy	>18 years	Recruiting	Dec 2023
NCT04128501	USA	Morphological remission post allo	II	Ven/Aza maintenance	18–75 years	Recruiting	Oct 2022
NCT03504644	USA	Relapsed/refractory	1B/II	Venetoclax + liposomal vincristine	>18 years	Recruiting	April 2021
New drug formulations							
NCT03575325	USA	Relapsed/refractory	II	Liposomal daunorubicin & cytarabine (CPX 351)	>18 years	Recruiting	Oct 2021
Immunotherapy							
NCT03081910	USA	Relapsed/refractory	I	CD5CAR/28zeta CAR T cells	<75 years	Recruiting	July 2021
NCT02742727	China	$CD7^{+}$ Relapsed/refractory	I/II	anti CD7 CAR pNK cells	>18 years	Unknown	March 2017

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

Table I. (*Continued*)

Trial number	Countries recruiting	Disease status	Phase	Treatment	Age	Current status	Est. closing date
NCT04033302	China	Unknown	I	Anti CD7 CAR T cells	6 months-75 years	Recruiting	July 2021
NCT03690011	USA	Relapsed/refractory	I	CD7CAR/28zeta CAR T cells	<75 years	Not yet recruiting	May 2023
NCT04004637	China	Relapsed/refractory	I	CD7 CAR T cells	7–70 years	Recruiting	June 2021
Monoclonal antibodies							
NCT03384654	USA/Germany/Belgium/ France/Israel/Italy/Netherlands/ Spain/Sweden/UK	Relapsed/refractory	II	Daratumumab + vinc/dox/pred/v daratumumab PEG asparaginase/ cyclophosphamide/ cytarabine/6MP/mtx	1–30 years	Recruiting	Dec 2021
Other							
NCT02553460	USA/Canada	New diagnosis	I/II	Bortezomib & vorinostat + standard care	<1 year	Recruiting	July 2021
NCT02112916	USA/Australia/Canada/ New Zealand	New diagnosis	III	Bortezomib + cyclophosphamide/ cytarabine/daunorubicin/ dexamethasone/doxorubicin/ etoposide/ifosfamide/leucovarin/ 6MP/PEGasparaginase/ methotrexate/thioguanine/vincristine	2–30 years	Active, not recruiting	March 2020
NCT02795520	USA	Relapsed refractory ALL/ AML/CML/Advanced MPN/Advanced MDS	I/II	OTS167IV (MELK inhibitor)	>18 years	Recruiting	Dec 2021
NCT02484430	USA	Relapsed/refractory	II	Sapanisertib (mTOR inhibitor)	>18 years	Active, not recruiting	May 2021
NCT02293109	USA	New diagnosis	I	Carfilzomib + hyper CVAD	18–64 years	Active, not recruiting	Nov 2020
NCT03110354	USA	Relapsed/refractory	I	DS-3201b (EZH1/2 inhibitor)	>18 years	Recruiting	May 2021
NCT02890758	USA	Relapsed/refractory	I	NK cells + ALT803 (IL15)	>18 years	Recruiting	Feb 2021
NCT02392572	USA	Relapsed/refractory	I/II	ONC201 (Apoptosis initiator) +/− LDAC	>18 years	Active, not recruiting	Nov 2022
NCT02663518	USA/Canada	Relapsed/refractory	I	TTI-621 (SIRPaFc blocker)	>18 years	Recruiting	June 2021
NCT04446130	China	Newly diagnosed ETP-ALL	III	Decitabine (+HAAG chemotherapy)	15–60 years	Recruiting	Jan 2023
NCT03553238	China	Newly diagnosed ETP-ALL	II/III	Chidamide (HDACi) + chemotherapy	14–55 years	Recruiting	May 2020
NCT03860844	Denmark/Finland/Norway/ Netherlands/Sweden	Relapsed/refractory inc AML	II	Isatuximab + standard chemotherapy	28 days–18 years	Recruiting	Aug 2021

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

negative NK effector cell avoids any problem with CAR fratricide. However, surface CD3 is a relatively mature T cell marker that would not be expressed in all T-ALL cases and this approach would again result in fatal T cell aplasia, albeit depending on the persistence of NK activity. Similarly an anti-CD5 CAR NK cell has been reported to show activity in a xenograft model.[117]

Intriguing results have been presented from an anti-CD5 CAR T cell.[118] CD5 is expressed on most normal T cells and IgM secreting innate B1 B cells. In response to the anti-CD5 CAR T cell both normal and malignant T cells down-regulate CD5, but normal T cells additionally up-regulate anti-apoptotic proteins including BCL2 and PI-9 protecting themselves from cell death. The result is ongoing anti-tumour activity, with minimal CAR T fratricide. Importantly, these results show that CAR T efficacy is not just limited to choice of antigenic target, but also the susceptibility of the target cells to the effector mechanisms of the CAR T cells. A potential problem with this approach, however, is that chronic exposure to the CD5 antigen may result in CAR T exhaustion.

Recent positive pre-clinical results have been achieved targeting the CD1a antigen expressed in cortical T-ALL.[119] However, $CD1a^+$ T-ALL has a good prognosis, meaning relapsed/refractory $CD1a^+$ T-ALL is rare.[120]

One approach to avoid CAR T cell fratricide is to delete the targeted antigen in the CAR T cells during CAR T cell production. This has been successfully achieved in an anti-CD7 CAR T cell that has its own *CD7* loci disrupted by CRISPR/Cas9 genome editing.[121] Importantly, these CAR T cells demonstrated both effective anti-tumour responses, but also the ability to respond to viral peptides indicating that some broader cellular immunity may be re-established from the CAR T population itself. Another recently reported strategy targeting CD7 involves a CD7 expression blocker which results in the intracellular retention of CD7. Eight patients treated with this approach had limited cytokine release syndrome (CRS) (including no reported neurotoxicity) and 50% remain cytokine release syndrome (CRS) MRD negative (Zhang M et al, ASH 05/12/20).

An elegant approach has sought to exploit the mutual exclusive expression of TRBC1 and TRBC2 at the *TCRβ* β-constant region, a process with similarities to B cell kappa/lambda restriction.[122] TRBC1 and TRBC2 differ by just four amino acids. Normal T cells express one or other, but not both, while clonal disorders such as T-ALL will express only one of the antigens Fig 6. Generating a CAR T cell against TRBC1 circumvents both CAR T cell fratricide and T cell aplasia (killing only 50% of polyclonal T-cells), while showing activity against TRBC1+ T-ALL cell lines in a xenograft model. Early phase clinical trials of this CAR T cell in T cell lymphoma are currently recruiting. It should be noted that high-risk ETP-ALL cases that have arrested prior to *TCRβ* rearrangement will not express either isoform, with only approximately 25% of T-ALL cases amenable to TRBC1 directed therapy.[122]

Considerations in multiply relapsed and refractory patients ineligible for clinical trials

If possible, all relapsed/refractory patients should be entered onto clinical trials (Table I). For refractory patients fit for intensive therapy, our standard practice has been to attempt re-induction with either Fludarabine-Cytarabine-Idarubicin (FLA-Ida) or a Nelarabine containing regimen. However, for patients ineligible for clinical trials who are fit for active treatment, we would recommend considering the following possibilities, although this assumes the ability to access the various

Table II. Potential drug treatments for consideration in relapsed/refractory T-ALL.

Drug	Adjuvant treatment	Appropriate patient cohort
Nelarabine	Can be used as single agent Combination with etoposide & cyclophosphamide (NEC protocol)	Caution in patients with neuropathy
Venetoclax	Vincristine/steroids/daunorubicin azacytidine/decitabine	ETP-ALL
Navitoclax	Vincristine/steroids/daunorubicin Azacytidine/decitabine in combination with venetoclax	Non-ETP-ALL Monitor for thrombocytopenia
Bortezomib	Vincristine/steroids/daunorubicin/asparaginase	Caution in patients with neuropathy Patients fit for re-induction
FLT3 inhibitor	Can be used as single agent	FLT3 mutated (approx. 5% T-ALL) Associated with ETP-ALL and $CD117^+$
Ruxolitinib (or other JAKi)	Dexamethasone (pre-clinical data suggesting synergy)	IL7R or JAK activating mutations
Dasatinib	Either as single agent or with chemotherapy combination	No current validated biomarkers for patient selection. LCK phosphorylation or in vitro drug sensitivity testing may be of value
Daratumumab	Vincristine/steroids/daunorubicin/asparaginase	Appropriate antigen expression. Clinical use currently unproven for brentuximab and gemtuzumab

 © 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

agents on compassionate access schemes or through local/personal funding streams, accepting use of many of these agents is outside licence, and usage should be assessed by a specialist team on a case-by-case basis (Table II). Genetic and drug profiling is likely to assist the best treatment approach.

Conclusion

Instigating novel therapies for T-ALL is challenging due to the rarity of the disease, genetic heterogeneity and the toxicity associated with ablating the T cell repertoire. Immunotherapy has an increasing role in haematological cancers, with CAR T cell trials beginning in T-lineage malignancies. However, CAR T therapy requires expensive infrastructural support, both in terms of production and delivery, and is unlikely to be available outside of select institutes in high-income countries. Advances in our understanding of the genetics and epigenetics of this disease have contributed to the novel uses of previously well-described therapies, as well as the advent of new drugs. Targeting synthetic lethal pathways, such as recently described for CHK1 inhibition in EZH2 deficient T-ALL, offers the opportunity to spare normal tissues and reduce toxicity.[123] Personalised medicine has already been demonstrated to identify new therapeutic targets in cancer, including T-ALL, and we strongly believe that this approach will transform the outlook for this disease.[124] We propose a combination of genomics and drug profiling may enable the most appropriate treatment to be selected. However, such an approach is not without its difficulties, given the cost and requirement for rapid results, especially in a disease as aggressive as T-ALL. The paucity of validated biomarkers means there is no quick answer for the best treatment for an individual and for the moment such approaches are only feasibly delivered by large research centres. Lastly, given the rarity of the disease, multinational collaborative trials will be required to make meaningful progress in the relapsed/refractory setting.

Acknowledgements

R. P. is supported by a clinician-scientist fellowship from the Kay Kendall Leukaemia Fund. S. R. is supported by a Clinician Scientist Fellowship from Cancer Research UK (C67279/A27957). N. F. was funded by Great Ormond Street Hospital Children's Charity and a Cancer Research UK Clinical PhD studentship. M. R. M. was funded by a Blood Cancer UK Bennett Fellowship and a Biomedical Research Council Fellowship. Research in the Wellcome - MRC Cambridge Stem Cell Institute is funded by a grant from the Wellcome Trust (203151/Z/16/Z).

Author Contributions

RP, SER, NF and MM wrote the manuscript and generated the figures. RP and MM had final responsibility to submit for publication.

Conflict of interest

The authors declare no competing interests.

References

1. Goulden NJ, Knechtli CJ, Garland RJ, Langlands K, Hancock JP, Potter MN, et al. Minimal residual disease analysis for the prediction of relapse in children with standard-risk acute lymphoblastic leukaemia. *Br J Haematol*. 1998;**100**(1):235–44.
2. O'Connor D, Enshaei A, Bartram J, Hancock J, Harrison CJ, Hough R, et al. Genotype-specific minimal residual disease interpretation improves stratification in pediatric acute lymphoblastic leukemia. *J Clin Oncol*. 2018;**36**(1):34–43.
3. Schrappe M, Valsecchi MG, Bartram CR, Schrauder A, Panzer-Grumayer R, Moricke A, et al. Late MRD response determines relapse risk overall and in subsets of childhood T-cell ALL: results of the AIEOP-BFM-ALL 2000 study. *Blood*. 2011;**118**(8):2077–84.
4. Coustan-Smith E, Mullighan CG, Onciu M, Behm FG, Raimondi SC, Pei D, et al. Early T-cell precursor leukaemia: a subtype of very high-risk acute lymphoblastic leukaemia. *Lancet Oncol*. 2009;**10**(2):147–56.
5. Conter V, Valsecchi MG, Buldini B, Parasole R, Locatelli F, Colombini A, et al. Early T-cell precursor acute lymphoblastic leukaemia in children treated in AIEOP centres with AIEOP-BFM protocols: a retrospective analysis. *Lancet Haematol*. 2016;**3**(2):e80–e86.
6. Patrick K, Wade R, Goulden N, Mitchell C, Moorman AV, Rowntree C, et al. Outcome for children and young people with Early T-cell precursor acute lymphoblastic leukaemia treated on a contemporary protocol, UKALL 2003. *Br J Haematol*. 2014;**166**(3):421–4.
7. Farah N, Kirkwood AA, Rahman S, Leon T, Jenkinson S, Gale RE, et al. Prognostic impact of the absence of biallelic deletion at the TRG locus for pediatric patients with T-cell acute lymphoblastic leukemia treated on the Medical Research Council UK Acute Lymphoblastic Leukemia 2003 trial. *Haematologica*. 2018;**103**(7):e288–e292.
8. Goldstone AH, Richards SM, Lazarus HM, Tallman MS, Buck G, Fielding AK, et al. In adults with standard-risk acute lymphoblastic leukemia, the greatest benefit is achieved from a matched sibling allogeneic transplantation in first complete remission, and an autologous transplantation is less effective than conventional consolidation/maintenance chemotherapy in all patients: final results of the International ALL Trial (MRC UKALL XII/ECOG E2993). *Blood*. 2008;**111**(4):1827–33.
9. Vora A, Goulden N, Wade R, Mitchell C, Hancock J, Hough R, et al. Treatment reduction for children and young adults with low-risk acute lymphoblastic leukaemia defined by minimal residual disease (UKALL 2003): a randomised controlled trial. *Lancet Oncol*. 2013;**14**(3):199–209.
10. Sive JI, Buck G, Fielding A, Lazarus HM, Litzow MR, Luger S, et al. Outcomes in older adults with acute lymphoblastic leukaemia (ALL): results from the international MRC UKALL XII/ECOG2993 trial. *Br J Haematol*. 2012;**157**(4):463–71.
11. Silverman LB, Gelber RD, Dalton VK, Asselin BL, Barr RD, Clavell LA, et al. Improved outcome for children with acute lymphoblastic leukemia: results of Dana-Farber Consortium Protocol 91–01. *Blood*. 2001;**97**(5):1211–8.
12. Schrappe M, Reiter A, Ludwig WD, Harbott J, Zimmermann M, Hiddemann W, et al. Improved outcome in childhood acute lymphoblastic leukemia despite reduced use of anthracyclines and cranial radiotherapy: results of trial ALL-BFM 90. German-Austrian-Swiss ALL-BFM Study Group. *Blood*. 2000;**95**(11):3310–22.
13. Boiers C, Richardson SE, Laycock E, Zriwil A, Turati VA, Brown J, et al. A human IPS model implicates embryonic B-myeloid fate restriction as developmental susceptibility to B acute lymphoblastic leukemia-associated ETV6-RUNX1. *Dev Cell*. 2018;**44**(3):362–77 e7.
14. De Bie J, Demeyer S, Alberti-Servera L, Geerdens E, Segers H, Broux M, et al. Single-cell sequencing reveals the origin and the order of mutation

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

acquisition in T-cell acute lymphoblastic leukemia. *Leukemia*. 2018;**32**(6):1358–69.
15. Szczepanski T, van der Velden VH, Waanders E, Kuiper RP, Van Vlierberghe P, Gruhn B, et al. Late recurrence of childhood T-cell acute lymphoblastic leukemia frequently represents a second leukemia rather than a relapse: first evidence for genetic predisposition. *J Clin Oncol*. 2011;**29**(12):1643–9.
16. Marks DI, Paietta EM, Moorman AV, Richards SM, Buck G, DeWald G, et al. T-cell acute lymphoblastic leukemia in adults: clinical features, immunophenotype, cytogenetics, and outcome from the large randomized prospective trial (UKALL XII/ECOG 2993). *Blood*. 2009;**114**(25):5136–45.
17. Gaynon PS, Harris RE, Altman AJ, Bostrom BC, Breneman JC, Hawks R, et al. Bone marrow transplantation versus prolonged intensive chemotherapy for children with acute lymphoblastic leukemia and an initial bone marrow relapse within 12 months of the completion of primary therapy: Children's Oncology Group study CCG-1941. *J Clin Oncol*. 2006;**24**(19):3150–6.
18. Ko RH, Ji L, Barnette P, Bostrom B, Hutchinson R, Raetz E, et al. Outcome of patients treated for relapsed or refractory acute lymphoblastic leukemia: a therapeutic advances in Childhood Leukemia Consortium study. *J Clin Oncol*. 2010;**28**(4):648–54.
19. Samra B, Alotaibi AS, Short NJ, Khoury JD, Ravandi F, Garris R, et al. Outcome of adults with relapsed/refractory T-cell acute lymphoblastic leukemia or lymphoblastic lymphoma. *Am J Hematol*. 2020;**95**(9):E245–E247.
20. van den Berg H, de Groot-Kruseman HA, Damen-Korbijn CM, de Bont ES, Schouten-van Meeteren AY, Hoogerbrugge PM. Outcome after first relapse in children with acute lymphoblastic leukemia: a report based on the Dutch Childhood Oncology Group (DCOG) relapse all 98 protocol. *Pediatr Blood Cancer*. 2011;**57**(2):210–6.
21. Jenkinson S, Koo K, Mansour MR, Goulden N, Vora A, Mitchell C, et al. Impact of NOTCH1/FBXW7 mutations on outcome in pediatric T-cell acute lymphoblastic leukemia patients treated on the MRC UKALL 2003 trial. *Leukemia*. 2013;**27**(1):41–7.
22. Asnafi V, Buzyn A, Le Noir S, Baleydier F, Simon A, Beldjord K, et al. NOTCH1/FBXW7 mutation identifies a large subgroup with favorable outcome in adult T-cell acute lymphoblastic leukemia (T-ALL): a Group for Research on Adult Acute Lymphoblastic Leukemia (GRAALL) study. *Blood*. 2009;**113**(17):3918–24.
23. Breit S, Stanulla M, Flohr T, Schrappe M, Ludwig WD, Tolle G, et al. Activating NOTCH1 mutations predict favorable early treatment response and long-term outcome in childhood precursor T-cell lymphoblastic leukemia. *Blood*. 2006;**108**(4):1151–7.
24. Jotta PY, Ganazza MA, Silva A, Viana MB, da Silva MJ, Zambaldi LJ, et al. Negative prognostic impact of PTEN mutation in pediatric T-cell acute lymphoblastic leukemia. *Leukemia*. 2010;**24**(1):239–42.
25. Remke M, Pfister S, Kox C, Toedt G, Becker N, Benner A, et al. High-resolution genomic profiling of childhood T-ALL reveals frequent copy-number alterations affecting the TGF-beta and PI3K-AKT pathways and deletions at 6q15-16.1 as a genomic marker for unfavorable early treatment response. *Blood*. 2009;**114**(5):1053–62.
26. Trinquand A, Tanguy-Schmidt A, Ben Abdelali R, Lambert J, Beldjord K, Lengline E, et al. Toward a NOTCH1/FBXW7/RAS/PTEN-based oncogenetic risk classification of adult T-cell acute lymphoblastic leukemia: a Group for Research in Adult Acute Lymphoblastic Leukemia study. *J Clin Oncol*. 2013;**31**(34):4333–42.
27. Jenkinson S, Kirkwood AA, Goulden N, Vora A, Linch DC, Gale RE. Impact of PTEN abnormalities on outcome in pediatric patients with T-cell acute lymphoblastic leukemia treated on the MRC UKALL2003 trial. *Leukemia*. 2016;**30**(1):39–47.
28. Liu Y, Easton J, Shao Y, Maciaszek J, Wang Z, Wilkinson MR, et al. The genomic landscape of pediatric and young adult T-lineage acute lymphoblastic leukemia. *Nat Genet*. 2017;**49**(8):1211–8.
29. Rothenburger T, McLaughlin KM, Herold T, Schneider C, Oellerich T, Rothweiler F, et al. SAMHD1 is a key regulator of the lineage-specific response of acute lymphoblastic leukaemias to nelarabine. *Commun Biol*. 2020;**3**(1):324.
30. Kurtzberg J, Ernst TJ, Keating MJ, Gandhi V, Hodge JP, Kisor DF, et al. Phase I study of 506U78 administered on a consecutive 5-day schedule in children and adults with refractory hematologic malignancies. *J Clin Oncol*. 2005;**23**(15):3396–403.
31. Berg SL, Blaney SM, Devidas M, Lampkin TA, Murgo A, Bernstein M, et al. Phase II study of nelarabine (compound 506U78) in children and young adults with refractory T-cell malignancies: a report from the Children's Oncology Group. *J Clin Oncol*. 2005;**23**(15):3376–82.
32. DeAngelo DJ, Yu D, Johnson JL, Coutre SE, Stone RM, Stopeck AT, et al. Nelarabine induces complete remissions in adults with relapsed or refractory T-lineage acute lymphoblastic leukemia or lymphoblastic lymphoma: Cancer and Leukemia Group B study 19801. *Blood*. 2007;**109**(12):5136–42.
33. Commander LA, Seif AE, Insogna IG, Rheingold SR. Salvage therapy with nelarabine, etoposide, and cyclophosphamide in relapsed/refractory paediatric T-cell lymphoblastic leukaemia and lymphoma. *Br J Haematol*. 2010;**150**(3):345–51.
34. Kimberly P, Dunsmore SW, Devidas M, Wood BL, Esiashvili N, Eisenberg N, et al. COG AALL0434: a randomized trial testing nelarabine in newly diagnosed T-cell malignancy. *J Clin Oncol*. 2018;**36**(15_-suppl):10500.
35. Dunsmore KP, Winter SS, Devidas M, Wood BL, Esiashvili N, Chen Z, et al. Children's Oncology Group AALL0434: a phase III randomized clinical trial testing nelarabine in newly diagnosed T-cell acute lymphoblastic leukemia. *J Clin Oncol*. 2020;**38**(28):3282–93.
36. Lobry C, Oh P, Mansour MR, Look AT, Aifantis I. Notch signaling: switching an oncogene to a tumor suppressor. *Blood*. 2014;**123**(16):2451–9.
37. Weng AP, Millholland JM, Yashiro-Ohtani Y, Arcangeli ML, Lau A, Wai C, et al. c-Myc is an important direct target of Notch1 in T-cell acute lymphoblastic leukaemia/lymphoma. *Genes Dev*. 2006;**20**(15):2096–109.
38. Mansour MR, Linch DC, Foroni L, Goldstone AH, Gale RE. High incidence of Notch-1 mutations in adult patients with T-cell acute lymphoblastic leukemia. *Leukemia*. 2006;**20**(3):537–9.
39. Deangelo DJ, Stone RM, Silverman LB, Stock W, Attar EC, Fearen I, et al. A phase I clinical trial of the notch inhibitor MK-0752 in patients with T-cell acute lymphoblastic leukemia/lymphoma (T-ALL) and other leukemias. *J Clin Oncol*. 2006;**24**(18):6585.
40. Zheng H, Pritchard DM, Yang X, Bennett E, Liu G, Liu C, et al. KLF4 gene expression is inhibited by the notch signaling pathway that controls goblet cell differentiation in mouse gastrointestinal tract. *Am J Physiol Gastrointest Liver Physiol*. 2009;**296**(3):G490–G498.
41. Wei P, Walls M, Qiu M, Ding R, Denlinger RH, Wong A, et al. Evaluation of selective gamma-secretase inhibitor PF-03084014 for its antitumor efficacy and gastrointestinal safety to guide optimal clinical trial design. *Mol Cancer Ther*. 2010;**9**(6):1618–28.
42. Real PJ, Tosello V, Palomero T, Castillo M, Hernando E, de Stanchina E, et al. Gamma-secretase inhibitors reverse glucocorticoid resistance in T cell acute lymphoblastic leukemia. *Nat Med*. 2009;**15**(1):50–8.
43. Knoechel B, Bhatt A, Pan L, Pedamallu CS, Severson E, Gutierrez A, et al. Complete hematologic response of early T-cell progenitor acute lymphoblastic leukemia to the γ-secretase inhibitor BMS-906024: genetic and epigenetic findings in an outlier case. *Cold Spring Harb Mol Case Stud*. 2015;**1**(1):a000539.
44. Borthakur G, Martinelli G, Raffoux E, Chevallier P, Chromik J, Lithio A, et al. Phase 1 study to evaluate Crenigacestat (LY3039478) in combination with dexamethasone in patients with T-cell acute lymphoblastic leukemia and lymphoma. *Cancer*. 2020;**127**(3):372–80.
45. Habets RA, de Bock CE, Serneels L, Lodewijckx I, Verbeke D, Nittner D, et al. Safe targeting of T cell acute lymphoblastic leukemia by pathology-specific NOTCH inhibition. *Sci Transl Med*. 2019;**11**(494): eaau6246.
46. Moellering RE, Cornejo M, Davis TN, Del Bianco C, Aster JC, Blacklow SC, et al. Direct inhibition of the NOTCH transcription factor complex. *Nature*. 2009;**462**(7270):182–8.
47. Koyama D, Kikuchi J, Hiraoka N, Wada T, Kurosawa H, Chiba S, et al. Proteasome inhibitors exert cytotoxicity and increase chemosensitivity via

 © 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

transcriptional repression of Notch1 in T-cell acute lymphoblastic leukemia. *Leukemia*. 2014;**28**(6):1216–26.
48. Bertaina A, Vinti L, Strocchio L, Gaspari S, Caruso R, Algeri M, et al. The combination of bortezomib with chemotherapy to treat relapsed/refractory acute lymphoblastic leukaemia of childhood. *Br J Haematol*. 2017;**176**(4):629–36.
49. Mansour MR, Duke V, Foroni L, Patel B, Allen CG, Ancliff PJ, et al. Notch-1 mutations are secondary events in some patients with T-cell acute lymphoblastic leukemia. *Clin Cancer Res*. 2007;**13**(23):6964–9.
50. Knoechel B, Roderick JE, Williamson KE, Zhu J, Lohr JG, Cotton MJ, et al. An epigenetic mechanism of resistance to targeted therapy in T cell acute lymphoblastic leukemia. *Nat Genet*. 2014;**46**(4):364–70.
51. O'Neil J, Grim J, Strack P, Rao S, Tibbitts D, Winter C, et al. FBW7 mutations in leukemic cells mediate NOTCH pathway activation and resistance to gamma-secretase inhibitors. *J Exp Med*. 2007;**204**(8):1813–24.
52. Palamarchuk A, Efanov A, Maximov V, Aqeilan RI, Croce CM, Pekarsky Y. Akt phosphorylates Tal1 oncoprotein and inhibits its repressor activity. *Cancer Res*. 2005;**65**(11):4515–9.
53. Gutierrez A, Sanda T, Grebliunaite R, Carracedo A, Salmena L, Ahn Y, et al. High frequency of PTEN, PI3K, and AKT abnormalities in T-cell acute lymphoblastic leukemia. *Blood*. 2009;**114**(3):647–50.
54. Bandapalli OR, Zimmermann M, Kox C, Stanulla M, Schrappe M, Ludwig WD, et al. NOTCH1 activation clinically antagonizes the unfavorable effect of PTEN inactivation in BFM-treated children with precursor T-cell acute lymphoblastic leukemia. *Haematologica*. 2013;**98**(6):928–36.
55. Piovan E, Yu J, Tosello V, Herranz D, Ambesi-Impiombato A, Da Silva AC, et al. Direct reversal of glucocorticoid resistance by AKT inhibition in acute lymphoblastic leukemia. *Cancer Cell*. 2013;**24**(6):766–76.
56. Shepherd C, Banerjee L, Cheung CW, Mansour MR, Jenkinson S, Gale RE, et al. PI3K/mTOR inhibition upregulates NOTCH-MYC signalling leading to an impaired cytotoxic response. *Leukemia*. 2013;**27**(3):650–60.
57. Daver N, Boumber Y, Kantarjian H, Ravandi F, Cortes J, Rytting ME, et al. A phase I/II study of the mTOR inhibitor everolimus in combination with HyperCVAD chemotherapy in patients with relapsed/refractory acute lymphoblastic leukemia. *Clin Cancer Res*. 2015;**21**(12):2704–14.
58. Hinton HJ, Alessi DR, Cantrell DA. The serine kinase phosphoinositide-dependent kinase 1 (PDK1) regulates T cell development. *Nat Immunol*. 2004;**5**(5):539–45.
59. Juntilla MM, Wofford JA, Birnbaum MJ, Rathmell JC, Koretzky GA. Akt1 and Akt2 are required for alphabeta thymocyte survival and differentiation. *Proc Natl Acad Sci USA*. 2007;**104**(29):12105–10.
60. Trinquand A, Dos Santos NR, Tran Quang C, Rocchetti F, Zaniboni B, Belhocine M, et al. Triggering the TCR developmental checkpoint activates a therapeutically targetable tumor suppressive pathway in T-cell leukemia. *Cancer Discov*. 2016;**6**(9):972–85.
61. Chatenoud L, Ferran C, Reuter A, Legendre C, Gevaert Y, Kreis H, et al. Systemic reaction to the anti-T-cell monoclonal antibody OKT3 in relation to serum levels of tumor necrosis factor and interferon-gamma [corrected]. *N Engl J Med*. 1989;**320**(21):1420–1.
62. Zenatti PP, Ribeiro D, Li W, Zuurbier L, Silva MC, Paganin M, et al. Oncogenic IL7R gain-of-function mutations in childhood T-cell acute lymphoblastic leukemia. *Nat Genet*. 2011;**43**(10):932–9.
63. Vicente C, Schwab C, Broux M, Geerdens E, Degryse S, Demeyer S, et al. Targeted sequencing identifies associations between IL7R-JAK mutations and epigenetic modulators in T-cell acute lymphoblastic leukemia. *Haematologica*. 2015;**100**(10):1301–10.
64. Shochat C, Tal N, Bandapalli OR, Palmi C, Ganmore I, te Kronnie G, et al. Gain-of-function mutations in interleukin-7 receptor-alpha (IL7R) in childhood acute lymphoblastic leukemias. *J Exp Med*. 2011;**208**(5):901–8.
65. Zhang J, Ding L, Holmfeldt L, Wu G, Heatley SL, Payne-Turner D, et al. The genetic basis of early T-cell precursor acute lymphoblastic leukaemia. *Nature*. 2012;**481**(7380):157–63.
66. Girardi T, Vicente C, Cools J, De Keersmaecker K. The genetics and molecular biology of T-ALL. *Blood*. 2017;**129**(9):1113–23.
67. Flex E, Petrangeli V, Stella L, Chiaretti S, Hornakova T, Knoops L, et al. Somatically acquired JAK1 mutations in adult acute lymphoblastic leukemia. *J Exp Med*. 2008;**205**(4):751–8.
68. Degryse S, De Bock CE, Cox L, Demeyer S, Gielen O, Mentens N, et al. JAK3 mutants transform hematopoietic cells through JAK1 activation, causing T-cell acute lymphoblastic leukemia in a mouse model. *Blood*. 2014;**124**(20):3092–100.
69. Maude SL, Dolai S, Delgado-Martin C, Vincent T, Robbins A, Selvanathan A, et al. Efficacy of JAK/STAT pathway inhibition in murine xenograft models of early T-cell precursor (ETP) acute lymphoblastic leukemia. *Blood*. 2015;**125**(11):1759–67.
70. Degryse S, Cools J. JAK kinase inhibitors for the treatment of acute lymphoblastic leukemia. *J Hematol Oncol*. 2015;**8**:91.
71. Mansour MR, Reed C, Eisenberg AR, Tseng JC, Twizere JC, Daakour S, et al. Targeting oncogenic Interleukein-7 receptor signaling with N-acetylcysteine in T-cell acute lymphoblastuc leukaemia. *Br J Haematol*. 2015;**168**(2):230–8.
72. Delgado-Martin C, Meyer LK, Huang BJ, Shimano KA, Zinter MS, Nguyen JV, et al. JAK/STAT pathway inhibition overcomes IL7-induced glucocorticoid resistance in a subset of human T-cell acute lymphoblastic leukemias. *Leukemia*. 2017;**31**(12):2568–76.
73. Verbeke D, Gielen O, Jacobs K, Boeckx N, De Keersmaecker K, Maertens J, et al. Ruxolitinib synergizes with dexamethasone for the treatment of T-cell acute lymphoblastic leukemia. *Hemasphere*. 2019;**3**(6):e310.
74. Li Y, Buijs-Gladdines JG, Cante-Barrett K, Stubbs AP, Vroegindeweij EM, Smits WK, et al. IL-7 receptor mutations and steroid resistance in pediatric T cell acute lymphoblastic leukemia: a genome sequencing study. *PLoS Medicine*. 2016;**13**(12):e1002200.
75. Springuel L, Hornakova T, Losdyck E, Lambert F, Leroy E, Constantinescu SN, et al. Cooperating JAK1 and JAK3 mutants increase resistance to JAK inhibitors. *Blood*. 2014;**124**(26):3924–31.
76. Tvorogov D, Thomas D, Liau NPD, Dottore M, Barry EF, Lathi M, et al. Accumulation of JAK activation loop phosphorylation is linked to type I JAK inhibitor withdrawal syndrome in myelofibrosis. *Sci Adv*. 2018;**4**(11):eaat3834.
77. De Smedt R, Morscio J, Reunes L, Roels J, Bardelli V, Lintermans B, et al. Targeting cytokine and therapy induced PIM1 activation in T-cell acute lymphoblastic leukemia and lymphoma. *Blood*. 2020;**135**(19):1685–95.
78. Chen L, Willis SN, Wei A, Smith BJ, Fletcher JI, Hinds MG, et al. Differential targeting of prosurvival Bcl-2 proteins by their BH3- only ligands allows complementary apoptotic function. *Mol Cell*. 2005;**17**(3):393–403.
79. Chonghaile TN, Roderick JE, Glenfield C, Ryan J, Sallan SE, Silverman LB, et al. Maturation stage of T-cell acute lymphoblastic leukemia determines BCL-2 versus BCL-XL dependence and sensitivity to ABT-199. *Cancer Discov*. 2014;**4**(9):1074–87.
80. Anderson NM, Harrold I, Mansour MR, Sanda T, McKeown M, Nagykary N, et al. BCL2-specific inhibitor ABT-199 synergizes strongly with cytarabine against the early immature LOUCY cell line but not more-differentiated T-ALL cell lines. *Leukemia*. 2014;**28**(5):1145–8.
81. Richard-Carpentier G, Jabbour E, Short NJ, Rausch CR, Savoy JM, Bose P, et al. Clinical experience with venetoclax combined with chemotherapy for relapsed or refractory T-cell acute lymphoblastic leukemia. *Clin Lymphoma Myeloma Leuk*. 2020;**20**(4):212–8.
82. Oltersdorf T, Elmore SW, Shoemaker AR, Armstrong RC, Augeri DJ, Belli BA, et al. An inhibitor of Bcl-2 family proteins induces regression of solid tumours. *Nature*. 2005;**435**(7042):677–81.
83. Roberts AW, Seymour JF, Brown JR, Wierda WG, Kipps TJ, Khaw SL, et al. Substantial susceptibility of chronic lymphocytic leukemia to BCL2 inhibition: results of a phase I study of navitoclax in patients with relapsed or refractory disease. *J Clin Oncol*. 2012;**30**(5):488–96.
84. Mansour MR, He S, Li Z, Lobbardi R, Abraham BJ, Hug C, et al. JDP2: an oncogenic bZIP transcription factor in T cell acute lymphoblastic leukemia. *J Exp Med*. 2018;**215**(7):1929–45.
85. Li Z, He S, Look AT. The MCL1-specific inhibitor S63845 acts synergistically with venetoclax/ABT-199 to induce apoptosis in T-cell acute lymphoblastic leukemia cells. *Leukemia*. 2019;**33**(1):262–6.

© 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.

86. Cidado J, Boiko S, Proia T, Ferguson D, Criscione SW, San Martin M, et al. AZD4573 is a highly selective CDK9 inhibitor that suppresses MCL-1 and induces apoptosis in hematologic cancer cells. *Clin Cancer Res*. 2020;**26**(4):922–34.
87. Leverson JD, Cojocari D. Hematologic tumor cell resistance to the BCL-2 inhibitor venetoclax: a product of its microenvironment? *Front Oncol*. 2018;**8**:458.
88. Barber KE, Martineau M, Harewood L, Stewart M, Cameron E, Strefford JC, et al. Amplification of the ABL gene in T-cell acute lymphoblastic leukaemia. *Leukemia*. 2004;**18**(6):1153–6.
89. Graux C, Cools J, Melotte C, Quentmeier H, Ferrando A, Levine R, et al. Fusion of NUP214 to ABL1 on amplified episomes in T-cell acute lymphoblastic leukaemia. *Nat Genet*. 2004;**36**(10):1084–9.
90. Quintas-Cardama A, Tong W, Manshouri T, Vega F, Lennon PA, Cools J, et al. Activity of tyrosine kinase inhibitors against human NUP214-ABL1-positive T cell malignancies. *Leukemia*. 2008;**22**(6):1117–24.
91. Vanden Bempt M, Demeyer S, Broux M, De Bie J, Bornschein S, Mentens N, et al. Cooperative enhancer activation by TLX1 and STAT5 drives development of NUP214-ABL1/TLX1-positive T cell acute lymphoblastic leukemia. *Cancer Cell*. 2018;**34**(2):271–85 e7.
92. Frismantas V, Dobay MP, Rinaldi A, Tchinda J, Dunn SH, Kunz J, et al. Ex vivo drug response profiling detects recurrent sensitivity patterns in drug-resistant acute lymphoblastic leukemia. *Blood*. 2017;**129**(11):e26–e37.
93. Deenik W, Beverloo HB, Wattel MM, van Esser JW, Valk PJ, Cornelissen JJ, et al. Rapid complete cytogenetic remission after upfront dasatinib monotherapy in a patient with a NUP214-ABL1-positive T-cell acute lymphoblastic leukemia. *Leukemia*. 2009;**23**(3):627–9.
94. Laukkanen S, Gronroos T, Polonen P, Kuusanmaki H, Mehtonen J, Cloos J, et al. In silico and preclinical drug screening identifies dasatinib as a targeted therapy for T-ALL. *Blood Cancer J*. 2017;**7**(9):e604.
95. Shi Y, Beckett MC, Blair HJ, Tirtakusuma R, Nakjang S, Enshaei A, et al. Phase II-like murine trial identifies synergy between dexamethasone and dasatinib in T-cell acute lymphoblastic leukemia. *Haematologica*. 2020. https://doi.org/10.3324/haematol.2019.241026.
96. Sicinska E, Aifantis I, Le Cam L, Swat W, Borowski C, Yu Q, et al. Requirement for cyclin D3 in lymphocyte development and T cell leukemias. *Cancer Cell*. 2003;**4**(6):451–61.
97. Choi YJ, Li X, Hydbring P, Sanda T, Stefano J, Christie AL, et al. The requirement for cyclin D function in tumor maintenance. *Cancer Cell*. 2012;**22**(4):438–51.
98. Pikman Y, Alexe G, Roti G, Conway AS, Furman A, Lee ES, et al. Synergistic drug combinations with a CDK4/6 inhibitor in T-cell acute lymphoblastic leukemia. *Clin Cancer Res*. 2017;**23**(4):1012–24.
99. Kwiatkowski N, Zhang T, Rahl PB, Abraham BJ, Reddy J, Ficarro SB, et al. Targeting transcription regulation in cancer with a covalent CDK7 inhibitor. *Nature*. 2014;**511**(7511):616–20.
100. Mansour MR, Abraham BJ, Anders L, Berezovskaya A, Gutierrez A, Durbin AD, et al. Oncogene regulation. An oncogenic super-enhancer formed through somatic mutation of a noncoding intergenic element. *Science*. 2014;**346**(6215):1373–7.
101. Olson CM, Jiang B, Erb MA, Liang Y, Doctor ZM, Zhang Z, et al. Pharmacological perturbation of CDK9 using selective CDK9 inhibition or degradation. *Nat Chem Biol*. 2018;**14**(2):163–70.
102. Burns MA, Liao ZW, Yamagata N, Pouliot GP, Stevenson KE, Neuberg DS, et al. Hedgehog pathway mutations drive oncogenic transformation in high-risk T-cell acute lymphoblastic leukemia. *Leukemia*. 2018;**32**(10):2126–37.
103. Dagklis A, Demeyer S, De Bie J, Radaelli E, Pauwels D, Degryse S, et al. Hedgehog pathway activation in T-cell acute lymphoblastic leukemia predicts response to SMO and GLI1 inhibitors. *Blood*. 2016;**128**(23):2642–54.
104. Vilas-Zornoza A, Agirre X, Abizanda G, Moreno C, Segura V, De Martino RA, et al. Preclinical activity of LBH589 alone or in combination with chemotherapy in a xenogeneic mouse model of human acute lymphoblastic leukemia. *Leukemia*. 2012;**26**(7):1517–26.
105. Waibel M, Vervoort SJ, Kong IY, Heinzel S, Ramsbottom KM, Martin BP, et al. Epigenetic targeting of Notch1-driven transcription using the HDACi panobinostat is a potential therapy against T-cell acute lymphoblastic leukemia. *Leukemia*. 2018;**32**(1):237–41.
106. Etchin J, Sanda T, Mansour MR, Kentsis A, Montero J, Le BT, et al. KPT-330 inhibitor of CRM1 (XPO1)-mediated nuclear export has selective anti-leukaemic activity in preclinical models of T-cell acute lymphoblastic leukaemia and acute myeloid leukaemia. *Br J Haematol*. 2013;**161**(1):117–27.
107. Sanda T, Tyner JW, Gutierrez A, Ngo VN, Glover J, Chang BH, et al. TYK2-STAT1-BCL2 pathway dependence in T-cell acute lymphoblastic leukemia. *Cancer Discov*. 2013;**3**(5):564–77.
108. Akahane K, Sanda T, Mansour MR, Radimerski T, DeAngelo DJ, Weinstock DM, et al. HSP90 inhibition leads to degradation of the TYK2 kinase and apoptotic cell death in T-cell acute lymphoblastic leukemia. *Leukemia*. 2016;**30**(1):219–28.
109. Wyatt KD, Bram RJ. Immunotherapy in pediatric B-cell acute lymphoblastic leukemia. *Hum Immunol*. 2019;**80**(6):400–8.
110. Zheng W, Medeiros LJ, Young KH, Goswami M, Powers L, Kantarjian HH, et al. CD30 expression in acute lymphoblastic leukemia as assessed by flow cytometry analysis. *Leuk Lymphoma*. 2014;**55**(3):624–7.
111. Bride KL, Vincent TL, Im SY, Aplenc R, Barrett DM, Carroll WL, et al. Preclinical efficacy of daratumumab in T-cell acute lymphoblastic leukemia (T-ALL). *Blood*. 2018;**131**(9):995–9.
112. Bonda A, Punatar S, Gokarn A, Mohite A, Shanmugam K, Nayak L, et al. Daratumumab at the frontiers of post-transplant refractory T-acute lymphoblastic leukemia-a worthwhile strategy? *Bone Marrow Transplant*. 2018;**53**(11):1487–9.
113. Ofran Y, Ringelstein-Harlev S, Slouzkey I, Zuckerman T, Yehudai-Ofir D, Henig I, et al. Daratumumab for eradication of minimal residual disease in high-risk advanced relapse of T-cell/CD19/CD22-negative acute lymphoblastic leukemia. *Leukemia*. 2020;**34**(1):293–5.
114. Akkapeddi P, Fragoso R, Hixon JA, Ramalho AS, Oliveira ML, Carvalho T, et al. A fully human anti-IL-7Rα antibody promotes antitumor activity against T-cell acute lymphoblastic leukemia. *Leukemia*. 2019;**33**(9):2155–68.
115. Pinz S, Unser S, Rascle A. Signal transducer and activator of transcription STAT5 is recruited to c-Myc super-enhancer. *BMC Mol Biol*. 2016;**17**:10.
116. Chen KH, Wada M, Firor AE, Pinz KG, Jares A, Liu H, et al. Novel anti-CD3 chimeric antigen receptor targeting of aggressive T cell malignancies. *Oncotarget*. 2016;**7**(35):56219–32.
117. Chen KH, Wada M, Pinz KG, Liu H, Lin KW, Jares A, et al. Preclinical targeting of aggressive T-cell malignancies using anti-CD5 chimeric antigen receptor. *Leukemia*. 2017;**31**(10):2151–60.
118. Mamonkin M, Rouce RH, Tashiro H, Brenner MK. A T-cell-directed chimeric antigen receptor for the selective treatment of T-cell malignancies. *Blood*. 2015;**126**(8):983–92.
119. Sanchez-Martinez D, Baroni ML, Gutierrez-Aguera F, Roca-Ho H, Blanch-Lombarte O, Gonzalez-Garcia S, et al. Fratricide-resistant CD1a-specific CAR T cells for the treatment of cortical T-cell acute lymphoblastic leukemia. *Blood*. 2019;**133**(21):2291–304.
120. Leong S, Inglott S, Papaleonidopoulou F, Orfinada K, Ancliff P, Bartram J, et al. CD1a is rarely expressed in pediatric or adult relapsed/refractory T-ALL: implications for immunotherapy. *Blood Adv*. 2020;**4**(19):4665–8.
121. Gomes-Silva D, Srinivasan M, Sharma S, Lee CM, Wagner DL, Davis TH, et al. CD7-edited T cells expressing a CD7-specific CAR for the therapy of T-cell malignancies. *Blood*. 2017;**130**(3):285–96.
122. Maciocia PM, Wawrzyniecka PA, Philip B, Ricciardelli I, Akarca AU, Onuoha SC, et al. Targeting the T cell receptor beta-chain constant region for immunotherapy of T cell malignancies. *Nat Med*. 2017;**23**(12):1416–23.
123. León TE, Rapoz-D'Silva T, Bertoli C, Rahman S, Magnussen M, Philip B, et al. EZH2-deficient T-cell acute lymphoblastic leukemia is sensitized to CHK1 inhibition through enhanced replication stress. *Cancer Discov*. 2020;**10**(7):998–1017.
124. Wong M, Mayoh C, Lau LMS, Khuong-Quang DA, Pinese M, Kumar A, et al. Whole genome, transcriptome and methylome profiling enhances actionable target discovery in high-risk pediatric cancer. *Nat Med*. 2020;**26**(11):1742–53.

 © 2021 The Authors. *British Journal of Haematology* published by British Society for Haematology and John Wiley & Sons Ltd.