Curiously, in null neutrophils only CD32-specific antibodies could block trogocytosis, suggesting that in the absence of FcγRIIIB, FcγRIIA can promote trogocytosis.

What is next? The immunologic activity of FcγRIIIA on neutrophils reported by Golay et al is quite remarkable in view of the relatively low expression of this receptor on the cells. There are ~200 000 FcγRIIIB and 20 000 to 40 000 FcγRIIA on neutrophils[2,4,10]; extrapolation based on the data reported by Golay et al suggests that the number of FcγRIIIA expressed on neutrophils may be a few thousand, yet substantial activity is demonstrable. To clearly examine the action of FcγRIIIA in the absence of high levels of FcγRIIIB, it will be necessary to examine neutrophils from other (rare) FcγRIIIB null donors. Alternatively, neutrophils of patients with paroxysmal nocturnal hemoglobinuria have about 10% of normal levels of FcγRIIIB, and enzymatic treatment of neutrophils with PI-phospholipase C leads to almost quantitative removal of FcγRIIIB with no effect on FcγRIIA.[2,3]

Experiments that evaluate the activity of FcγRIIIA on neutrophils should focus on mechanisms of activation on the basis of assays for γ-chain phosphorylation as well as for downstream signaling.[4] On the basis of the activity exhibited by FcγRIIIA on neutrophils, strategies that increase its expression might provide a reasonable approach to increasing defenses against infection. It might also be informative to revisit the synergy of CR3 with FcγR and determine the relative contribution of FcγRIIIA and FcγRIIIB to this phenomenon. Golay et al recognized that their findings may also pertain to the action of neutrophils in autoimmune disease. Both basic scientists and clinicians can look forward to additional progress in several areas of investigation on the basis of the article by Golay et al.

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CLINICAL TRIALS AND OBSERVATIONS

Comment on Huls et al, page 1457

Maintenance therapy for AML: are we there yet?

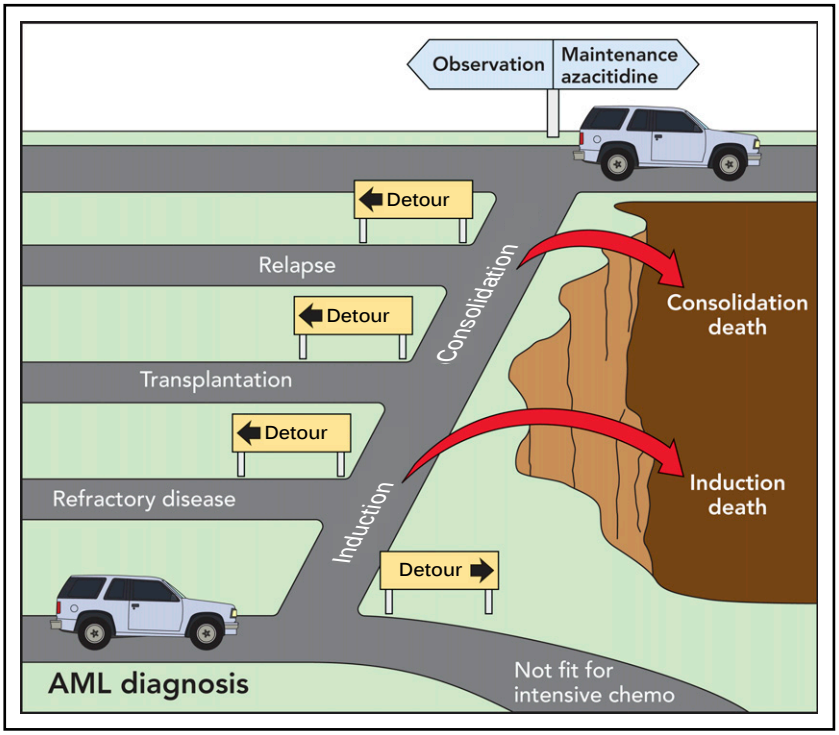
Andrew H. Wei | The Alfred Hospital; Monash University

In this issue of *Blood*, Huls et al present positive results from a randomized study (HOVON97) showing that disease-free survival (DFS) in older patients with acute myeloid leukemia (AML) was improved by azacitidine compared with postremission observation.[1]

Relapse after intensive induction and consolidation therapy remains the most important cause of treatment failure in AML. For older patients, the risk of relapse after intensive chemotherapy is 50% to 80%.[2] Allogeneic hematopoietic cell transplantation (allo-HCT) is an accepted option for select older patients with adverse cytogenetic and molecular risk factors. The role of maintenance therapy in reducing relapse risk in patients who do not receive transplants remains controversial.

Prior randomized studies have suggested clinical benefit for several maintenance strategies, including low-dose cytarabine, recombinant interleukin 2/histamine dihydrochloride, or attenuated chemotherapy (reviewed in Rashidi et al[3]). Many of these studies suffered from small sample size and concerns that the first-line chemotherapy was suboptimal by today's standards. A French study conducted in older patients demonstrated a delayed survival benefit for patients receiving 2 years of maintenance therapy with norethandrolone, an androgen analog.[4] The uncertain mechanism of action and limited norethandrolone availability worldwide have limited broad adoption and further exploration of this strategy. FLT3 inhibitors have also been actively explored in maintenance for patients with *FLT3*-mutant AML, with evidence strongest for their use in the postallograft setting.[3,5]

Recruitment of patients to maintenance studies may be challenging. A UK National Cancer Research Institute (NCRI) study found that only 28% of older patients initially registered to receive intensive chemotherapy (in first remission to maintenance azacitidine or not) underwent randomization.[6] This is typical of most maintenance studies, where enrolled patients represent a small fraction of the starting population. Patients need to survive intensive chemotherapy, achieve and remain in remission, and not be candidates for allo-HCT (see figure). Parenteral



The hazardous and detour-laden road to maintenance therapy in AML. chemo, chemotherapy. Professional illustration by Patrick Lane, ScEYEnce Studios.

drug administration and concerns regarding potential drug toxicity may represent further deterrents to maintenance trial participation.

Against this backdrop, the results from the HOVON97 trial are intriguing. The study randomized 116 eligible patients 60 years of age or older in complete remission (CR) or CR with incomplete hematologic recovery (CRi) after 2 cycles of intensive chemotherapy. Recruitment took 6.5 years, more than double the planned accrual time of 3 years, resulting in termination of the study prior to the planned recruitment target. Despite this, the primary end point was met, with DFS significantly improved from 10.3 to 15.9 months in the azacitidine arm. Treatment appeared tolerable, with almost two-thirds completing the 12 cycles of protocol treatment. Post hoc analyses suggested that azacitidine increased DFS for patients in CR or with a baseline platelet count of $\geq 100 \times 10^9$/L. In contrast, a benefit for azacitidine was not apparent for patients in CRi or with a platelet count of $<100 \times 10^9$/L. These results suggest the possibility that maintenance azacitidine may have the optimal effect in patients with higher-quality potentially measurable residual disease–negative (MRD^-) disease (see below).

In support of this hypothesis, the UK NCRI has presented preliminary data on the role of azacitidine maintenance in patients older than 60 years of age with AML in CR after 2 courses of intensive chemotherapy.[6] They found a significant survival benefit for azacitidine maintenance among patients without detectable measurable residual disease by flow cytometry (MRD^-), whereas a benefit for azacitidine was not observed in MRD^+ patients, suggesting that maintenance therapy had the highest utility in patients with chemosensitive disease. This raises the question of whether MRD^+ patients should instead be considered for alternative salvage approaches.

Will the HOVON97 study establish azacitidine maintenance as the standard of care for AML? The study certainly indicates the likelihood of a clinical effect from azacitidine in the postremission phase of therapy. Although maintenance was administered for a maximum of 12 cycles, it is unknown whether a longer period of maintenance treatment could have further improved DFS. In addition, overall survival (OS) was not significantly improved, although the study was not powered to show superiority for this end point. It is likely, however, that another confirmatory study using a more convenient azacitidine-dosing formulation on the back of this study could provide strong support for maintenance therapy as a standard approach for this group of elderly AML patients.

Indeed, a phase 3 randomized maintenance study (QUAZAR) has completed accrual, randomizing over 460 patients to 24 months of CC-486 (Celgene Corporation), an oral azacitidine analog vs placebo, in patients 55 years of age or older with AML in first CR or CRi after intensive chemotherapy.[7] The primary end point was OS. If this study is positive, it is likely that maintenance therapy will be accepted as a new standard of care for patients with AML. The HOVON97 study represents an important contribution to the positive potential of postremission maintenance therapy in AML. Remission induction and consolidation should now be considered the first step of a patient's AML journey. Next-generation sequencing (NGS) panels demonstrate persistence of recognized AML mutations in ~30% of patients in CR after intensive induction chemotherapy.[8] This excludes age-related *DNMT3A*, *TET2*, and *ASXL1* mutations, whose long-term relevance as markers of leukemic relapse remains uncertain. When NGS was combined with flow cytometry, MRD detected by either or both techniques was present in ~40% of patients and associated with relapse in 50% to 73% of cases, compared with 27% among patients without MRD. With standardized guidelines now available for the measurement and definition of MRD, the future incorporation of MRD monitoring and guided intervention during the postremission phase of AML is now a real possibility.[9] It is likely that postremission therapies could be further risk-adapted to incorporate or combine more target-directed options in the future, with FLT3 and isocitrate dehydrogenase inhibitors obvious candidates. With promising results from multicycle low-intensity treatment options in combination with the targeted B-cell lymphoma 2 inhibitor venetoclax reported, the distinction between induction, consolidation, and maintenance phases of treatment are already starting to blur.[10] The concept of maintenance therapy is moving from one of clinical uncertainty to one of clinical necessity. Therefore, for maintenance therapy in AML, although we

are not quite "there yet," we are certainly getting very close.

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MYELOID NEOPLASIA

Comment on Esnault et al, page 1495

Arsenic and old *FLT3*

Mark Levis | Johns Hopkins University

***FLT3*-internal tandem duplication (*FLT3*-ITD) mutations are common in acute promyelocytic leukemia (APL) and render the disease more difficult to manage or cure. In this issue of *Blood*, Esnault et al begin to unravel how the mutation-activated FLT3 receptor impedes the effects of all-*trans* retinoic acid (ATRA) and how arsenic counters this.[1]**

FLT3 activating mutations have thus far defied formal classification as an acute myeloid leukemia (AML) subtype. "FLT3-mutant AML" is not a World Health Organization–designated category of the disease, and for good reason. These mutations seem to delight in showing up in different subtypes of AML and making a bad situation worse. Take APL for example. FLT3-ITD mutations are found in roughly one-third of all patients with APL, and in the pre-arsenic era, they rendered APL more problematic to manage and adversely affected the survival of patients with an otherwise curable disease.[2,3]

Esnault et al crossed an FLT3-ITD knock-in transgenic mouse line with an established promyelocytic leukemia/retinoic acid receptor alpha (PML/RARA) mouse line to generate the double-mutant murine APL. There was no obvious difference in phenotype between the mice with PML/RARA and those with PML/RARA/FLT3-ITD, but the double-mutant mice showed a blunted response to ATRA, both in differentiation and in survival. The reorganization of nuclear bodies that would normally occur in response to ATRA did not occur in the double-mutant mice nor was PML/RARA degraded. The use of arsenic, however, induced differentiation, RARA degradation, and p53 induction in both models.

This demonstration that arsenic abrogates the FLT3-ITD–conferred resistance to ATRA makes sense in light of the clinical data. Shortly after they were discovered, FLT3-ITD mutations were noted to occur frequently in APL and were typically associated with a high white blood cell count at presentation, defining such patients as high risk by conventional criteria.[2] Treatment of FLT3-ITD–mutated APL with ATRA combined with chemotherapy was often successful but still resulted in worse overall survival compared with APL patients lacking such mutations.[3] The introduction of arsenic into the management of APL has led to remarkable survival rates,[4] but high-risk patients continue to be somewhat problematic. Interestingly, the findings from one clinical study are particularly salient to this topic.[5] Iland et al reported that high-risk APL patients treated with a regimen using both ATRA and arsenic fared slightly worse than low- or intermediate-risk patients but not if they had an FLT3 mutation. In other words, arsenic seemed to abrogate the prognostic effects of FLT3 mutations specifically. The findings of Esnault et al, therefore, provide important scientific support for the clinical results reported by Iland et al[5] and suggest that APL patients with FLT3 mutations will benefit from treatment with induction regimens incorporating both ATRA and arsenic, and they probably do not need an FLT3 inhibitor. For practical purposes, this essentially means that all APL patients should have arsenic as a component of induction, not just those with low- and intermediate-risk disease.

There is, perhaps, a larger story in all of this. Work over the past several years has yielded a model of AML in which the disease develops from the sequential acquisition of driver mutations.[6,7] In general, any individual driver mutation is insufficient to cause the disease. Likewise, targeting any individual driver mutation with single-agent therapy (such as with ATRA alone, an FLT3 inhibitor alone, or an IDH inhibitor alone) is insufficient to cure the disease. APL is the first subtype of AML that can be consistently cured (provided the patient gets to the hospital in time). However, to accomplish that, it seems that the mutations driving each particular case have to be dealt with in some way. For the other AML subtypes driven by multiple different mutations, we can expect the same.