

## BRIEF REPORT

# Clinical Application of Asparaginase Activity Levels Following Treatment With Pegaspargase

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Asparaginase, an enzyme used to treat acute lymphoblastic leukemia and related forms of nonHodgkin lymphoma, depletes asparagine, which leads to lymphoblast cell death. Unlike most chemotherapeutic agents, asparaginase is a foreign protein that can result in clinical allergy and/or silent hypersensitivity with production of neutralizing antibodies that inactivate asparaginase. In North America, asparaginase activity levels can now be obtained via a

commercially available assay, for therapeutic drug monitoring and investigation of potential allergic reactions. Herein, we provide recommendations and a corresponding algorithm for the clinical application of this assay after treatment with pegaspargase to evaluate suspected hypersensitivity reactions and/or silent inactivation. *Pediatr Blood Cancer* © 2014 Wiley Periodicals, Inc.

**Key words:** ALL; cancer pharmacology; chemotherapy; lymphoblastic lymphoma; pharmacokinetics

## INTRODUCTION

The clinical effect of asparaginase (ASNase) is related to depletion of serum asparagine (ASN), an essential amino acid for acute lymphoblastic leukemia (ALL) cells [1]. Measuring ASN in blood samples is challenging because ASNase continues to degrade ASN *ex vivo*, resulting in falsely low levels of ASN. Thus, ASNase enzyme activity is used as a surrogate for ASN depletion [1–6]. Recently, a commercial ASNase assay became available in the U.S. [7]. Other assays available in the U.S. on a research basis are not approved for commercial use. In other countries research-based assays can be used in clinical settings [8,9], such as the Medac Asparaginase Activity Test [10].

In this report, we offer recommendations for use of the U.S. commercial assay following treatment with pegylated (polyethylene glycol) *Escherichia coli* ASNase (pegaspargase, pegASNase), the primary first-line agent, that are summarized in an algorithm (Fig. 1). Recommendations for monitoring native *E. coli* ASNase and *Erwinia* ASNase activity are not provided because in the U.S. the former is no longer available and the latter is used almost exclusively following pegASNase allergy [11] and when it can no longer be used, there are no alternatives.

## METHODS

The algorithm serves three basic functions: (i) *Clinical Reaction*: To assess whether or not a suspected reaction is associated with neutralizing antibody (NA) formation that results in inadequate ASNase activity (and ASN depletion), the algorithm checks for a lower-than-expected ASNase activity shortly after the clinical reaction; (ii) *Possible Silent Inactivation*: To detect NA without clinical symptoms, the algorithm tests for a very low ASNase activity (<0.05 IU/mL) 4–7 days after standard-dose pegASNase; and (iii) *Possible Accelerated Terminal-Phase Clearance*: Some patients have adequate ASNase activity for a week or more and then rapidly disappear. The algorithm compares ASNase activity level 7 days after an adequate first level.

When to measure serum enzyme activity after pegASNase administration depends on the indication and the amount of pegASNase received if less than full dose was administered. To assess a *Clinical Reaction* or *Possible Silent Inactivation*, the level should be obtained before enzyme activity becomes too low to

measure accurately or too late to optimally replace an ineffective dose. The algorithm was based on enzyme activity measured with the new assay on Children's Oncology Group studies and on pegASNase's known primary plasma half-life of  $5.7 \pm 3.2$  days [12]. During the terminal phase of clearance, the half-life normally shortens with time [13,14].

## RESULTS

### Suspected Allergic Reaction (*Clinical Reaction*)

Many allergic reactions to ASNase are classic acute hypersensitivity phenomena with wheezing, dyspnea, angioedema (Fig. 2), or diffuse urticarial rash. However, there are also uncertain symptomatic reactions, for which pegASNase could be administered again if there is confidence of clinical efficacy.

Since an ASNase activity of  $\geq 0.05$  IU/mL is considered therapeutic [15], an ASNase activity of <0.05 IU/mL prior to/on day 7 after a full dose suggests concomitant NA and a switch to *Erwinia* ASNase. The sample should thereby be collected when ASNase activity is expected to be  $\gg 0.05$  IU/ml (Fig. 1), which for practical purpose is assessed 4–7 days after a full-dose of pegASNase. If the reaction occurs during the infusion and less than half of the drug is given, the algorithm provides for earlier sampling depending on how much of the dose was administered.

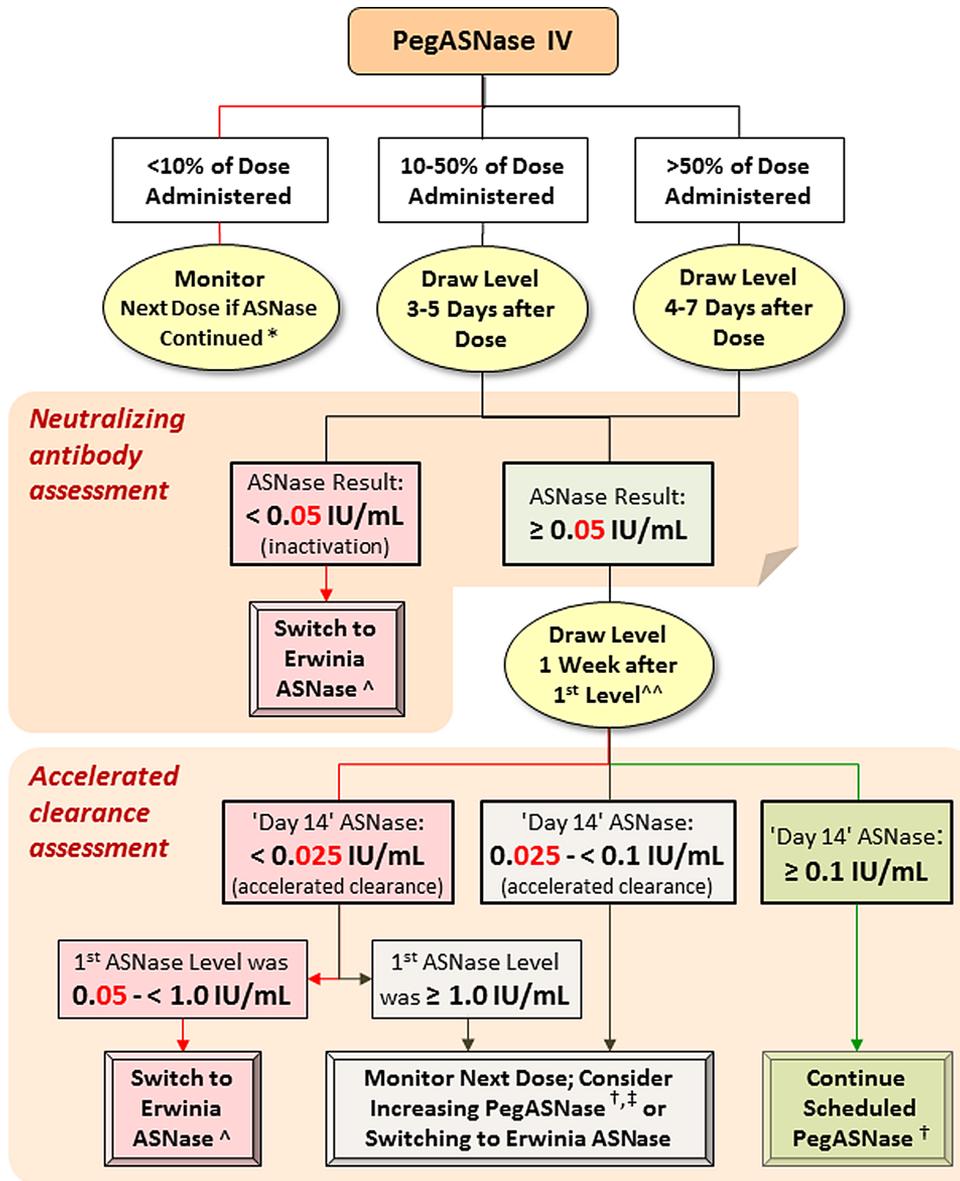
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\* In most instances, the ASNase preparation would be discontinued  
 ^^ 7 days after 1st sample  
 ^ 6 doses of 25,000 IU/m<sup>2</sup> IM at 2-3 day intervals for each scheduled dose of pegASNase  
 † With premedication to prevent a clinical allergic reaction if ASNase levels monitored  
 ‡ Administering another dose of pegASNase should be considered,<sup>17</sup> as well as monitoring future doses levels and continued accelerated terminal-phase disappearance

**Fig. 1.** Algorithm for pegASNase activity level determination after clinical allergic reaction and to assess for possible silent inactivation by neutralizing antibody and/or accelerated terminal-phase clearance.

In the case of non-life-threatening reactions and continuation of the ASNase therapy is considered, monitoring ASNase activity can guide additional pegASNase therapy to compensate for the missed or abbreviated dose. If the level is  $\geq 0.05$  IU/mL, it may be reasonable to continue pegASNase along with anti-allergy premedication such as hydrocortisone, diphenhydramine and acetaminophen, as illustrated by a recent patient.

**CASE DESCRIPTION**

A 15-year-old male with relapsed ALL had previously received numerous doses of pegASNase. While receiving pegASNase after high-dose cytarabine, he complained of difficulty breathing but was not wheezing and had normal oxygen saturation. Four hours following completion of the pegASNase infusion, he developed an



**Fig. 2.** Patient with allergic reaction to asparaginase. *Ethics Statement:* Assent from the patient and informed consent from his mother were obtained for publication of this photograph.

urticarial rash over his arms and trunk and complained of chest tightness and discomfort with breathing. Diphenhydramine and hydrocortisone were administered and the symptoms resolved. Because of the late onset of symptoms and uncertainty about a true allergic reaction, a serum ASNase activity level was obtained 96 hr post infusion. Based on a result of 0.87 IU/ml that excluded NA, he continued on intravenous pegASNase, receiving premedication with subsequent infusions and albeit with some vague clinical symptoms was able to tolerate continued pegASNase therapy.

### Subclinical Presence of ASNase NA After Prior Exposure (*Possible Silent Inactivation*)

A common scenario is a patient with relapsed leukemia who was previously exposed to ASNase, does not have a history of ASNase allergy, and may benefit from continued ASNase. The concern is that the patient may have previously developed NA, which *silently* inactivates the ASNase. This is most likely to occur in a patient whose prior therapy included native ASNase since it is more immunogenic than pegASNase. Silent NA may also be considered if the leukemia is responding too slowly during an ASNase-containing therapy phase. A low ASNase activity level may be overcome by administering the pegASNase more frequently [16], or by switching to *Erwinia* ASNase. Either way, the algorithm (Fig. 1) can be used to guide future therapy. If there is suspicion for NA formation, the ASNase activity level should be assessed after the second administration since it takes the first exposure to induce antibody formation.

Administering anti-allergy medications prior to ASNase doses can also be applied as a prevention strategy. In the U.S. adult-intergroup C10403 trial, an amendment that required premedication with hydrocortisone, diphenhydramine, and acetaminophen before

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each ASNase dose resulted in a reduction in grade 3–4 hypersensitivity reactions from 12.9% to 7.9% [17]. The concern that this approach may mask the presence of NA and thereby enable silent inactivation can be addressed by measuring ASNase activity in such patients according to the algorithm.

### More Rapid ASNase Clearance (*Possible Accelerated Clearance*)

Some patients have adequate ASNase activity for the first week but thereafter have lower-than-expected levels suggesting accelerated terminal-phase clearance and a shortened interval of asparagine depletion. The algorithm thereby also recommends comparing a second ASNase level 1 week after the first one. If the difference suggests accelerated clearance, interpreted according to the amount of intended dose delivered, recommended options include repeating a dose of pegASNase or switching to *Erwinia* ASNase, and in either case monitoring ASNase activity after next doses.

## DISCUSSION

The pegASNase algorithm should help identify patients who need not discontinue pegASNase therapy, the rare patient with silent NA who should be switched to *Erwinia* ASNase, and patients with accelerated terminal-phase clearance that merits additional pegASNase administration. If *Erwinia* ASNase is substituted for pegASNase, six doses of 25,000 IU/m<sup>2</sup> should be administered (intramuscularly for U.S. patients) at 2- to 3-day intervals to replace each scheduled dose of pegASNase, including as appropriate the ineffective dose.

Accentuation of the normal accelerated terminal-phase of clearance is a new observation. It is unlikely due to NA, since these patients have complete ASNase depletion for a week or more. It may be due to other non-NAs that increase enzyme-antibody complex clearance.

The algorithm was not designed to address other potential indications such as trough monitoring to individualize dosing [18,19] and overcoming NA with desensitization [20] and/or increasing dose intensity. As experience with the algorithm accrues, it will undoubtedly be improved and may ultimately acquire other applications.

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