Abstract

Purpose. To review the literature examining the mixing of insulin glargine with rapid-acting insulin (RAI).

Methods. A literature search was conducted via PubMed and Medline (from 1948 to August 2012) using the search terms “diabetes,” “insulin glargine,” “short acting insulin,” “rapid acting insulin,” and “mixing.” Literature was limited to English-language articles reporting on human studies. Studies with data describing mixing glargine with any short-acting insulin or RAI were included. Four studies met inclusion criteria.

Results. Of the four studies assessing mixing glargine, one was a pharmacokinetic study. The other three assessed clinical outcomes in “real-world” settings. All of these studies were conducted in pediatric patients with type 1 diabetes. Two of the clinical outcomes studies did not report significant differences in A1C levels or preprandial, postprandial, or nocturnal blood glucose levels from mixing glargine and RAI. One of the clinical outcome studies reported improved blood glucose control (A1C and fasting blood glucose) with RAI mixed with glargine compared to RAI mixed with NPH insulin. There were no significant differences in hypoglycemia in any of the clinical outcome trials at any time measured.

Conclusion. Initial small clinical trials indicate that there are no significant changes in clinical outcomes (blood glucose levels, A1C levels, and hypoglycemia) when mixing glargine with RAI. Additional studies with larger patient populations and longer trial durations are needed before mixing glargine with RAI can be recommended on a routine basis in clinical practice.

Type 1 diabetes accounts for about 5–10% of patients with diabetes; however, 75% of patients diagnosed with type 1 diabetes are < 18 years of age.1 The benefits of intense insulin therapy to lower A1C and prevent diabetes-related complications are well documented.

Insulin glargine, the first once-daily basal insulin approved by the U.S. Food and Drug Administration (FDA), is used in regimens to treat patients with type 1 or type 2 diabetes.2 The clear, colorless solution of glargine is buffered to a pH of 4 and is completely soluble at this level. After injection, a microprecipitate forms at the injection site that delays and prolongs glargine’s action. The FDA warns against mixing glargine with other insulins because of its pH and solubility.2 Therefore, basal-bolus therapy with glargine increases the number of daily injections compared to regimens using intermediate-acting NPH insulin. However, glargine offers less hypoglycemia than NPH because it is relatively peakless.3 Glargine is, however, more expensive than NPH.

The treatment of diabetes in children presents a number of challenges, including pain at injection sites, frequent injections, injection-related anxiety (pain or needle phobia), fear of hypoglycemia, and concerns about weight gain. The insulin regimen must be acceptable to patients
and their family, while achieving glycemic control and minimizing hypoglycemia. In patients 8–21 years of age, compliance decreases with frequency of injections. Some clinicians try to minimize the number of injections by mixing glargine with a rapid-acting insulin (RAI) despite FDA warnings against this practice. This review examines the data on mixing glargine with an RAI.

Methodology
A literature search was conducted via PubMed and Medline (articles published from 1948 to January 2012) using the search terms “diabetes,” “insulin glargine,” “short-acting insulin,” “rapid-acting insulin,” and “mixing.” The search was limited to articles published in English describing human studies. Studies with data describing mixing glargine with any short-acting insulin or RAI were included. Four studies met these inclusion criteria.

Literature Review
Four trials have been published regarding the mixing of insulin glargine and an RAI. Cengiz et al. conducted a glucose clamp study in 11 adolescents (six males and five females, mean age 15.1 ± 3 years) with type 1 diabetes. This trial compared the pharmacodynamic and pharmacokinetic effects of administering glargine mixed with insulin lispro to administering glargine separately. Patients had diabetes for at least 1 year and were on continuous subcutaneous insulin infusion for at least 3 months. The participants were admitted to the hospital the night before the study and randomized to receive 0.2 units/kg of lispro and 0.4 units/kg of glargine, either mixed or not mixed. The insulins were mixed at room temperature immediately before the injection. The insulin pump was suspended before the dose was given. Two clamp studies were performed, with patients receiving the opposite treatment regimen during the second study, which was within 4 weeks of the first clamp study.

Both the pharmacodynamic and pharmacokinetic profiles were altered with the mixing of glargine and lispro. The peak effect of lispro was significantly reduced in the mixing group (separate 7.1 ± 1 vs. mixed 3.9 ± 1 mg/kg/minute, P = 0.04). With regard to pharmacokinetics, the peak concentration of lispro was significantly less and the time to peak action was significantly delayed in the mixing group (Table 1). Mixing the two insulins flattens and delays the lispro peak. These results might lead to poorer control of postmeal glucose levels.

One limitation of this study is the use of a fixed 1:2 ratio of lispro to glargine. The insulin lispro dose could be increased to overcome the delay and diminished action peak. The delay of action with the lispro can be > 5 hours. Clinically, there could be an increased risk of nocturnal hypoglycemia if lispro and glargine were dosed at supper.

The results in this study are consistent with the results observed in previous animal studies when mixing regular insulin with glargine (delayed onset of action and a delayed time to maximum effect).\(^5\)

Kaplan et al. conducted a 30-day crossover study in 14 pediatric patients (6 male; mean age 13.5 ± 0.5 years) with type 1 diabetes. This trial compared the efficacy of mixing glargine with either lispro or aspart insulin and administering the doses twice daily compared to once daily. Continuous glucose monitoring (CGM) was used to assess each regimen. Patients used glargine at baseline once daily with three to four injections of an RAI. Patients were randomized to one of two groups with a crossover design: 1) separate injections (glargine before breakfast and before supper and RAI with each meal, and 2) mixed (pre-breakfast and pre-supper glargine mixed with an RAI and an RAI dose at lunch). Each patient underwent three studies over a 4- to 6-week period. Patients continued each regimen for 10 days, and CGM was performed during the final 3 days of each regimen.

There was no significant difference in hypoglycemia, preprandial glucose, or postprandial glucose levels between baseline, separate, and mixed groups (Table 1). However, the mixed group experienced lower nocturnal blood glucose levels, and the authors cautioned that titration of the evening dose of glargine may be required to prevent hypoglycemia.

This study was conducted under real-life conditions, and there was no difference in pain or reported reactions with the mixed regimen. The study did document cloudiness when glargine was mixed with lispro or aspart, which is consistent with data on file with the manufacturer regarding glargine mixing. Limitations of this study include a lack of discussion about the actual insulin doses and the short study duration (10 days for each study group).

Fiallo-Scharer et al. conducted a 24-week prospective, case-control study in 110 pediatric patients (70 male; mean age 13.4 ± 3.8 years in the mixed group and 12.9 ± 4.0 years in the nonmixed group) with type 1 diabetes. This trial compared the efficacy and safety of administering glargine mixed with either lispro or aspart to that of glargine administered separately from an RAI. The patients mixing insulin (n = 55) were matched with controls (n = 55) based on age, sex, duration of diabetes, and use of identical types of insulin but who were not mixing before administration. Patients were allowed to continue the doses and frequency of insulin administration they had been using at the time of enrollment for both glargine and their RAI. Data were collected for 6 months before and 6 months after enrollment. The primary outcome was change in A1C.

There was no significant difference between the two groups in A1C at 6 months (mixing 8.54 ± 1.14 vs. nonmixing 8.61 ± 1.14%, P = 1.0). There also were no significant differences in the incidence of nonsevere hypoglycemia, severe hypoglycemia, or diabetic ketoacidosis at 3 or 6 months (Table 1). However, it should be noted that four patients in the mixing group chose to discontinue mixing because of an increased frequency of hypo- and hyperglycemic episodes. The strengths of this trial included a well-matched controlled group and a 6-month follow-up period. Limitations included the study design, lack of blinding, lack of standardized insulin dosing and frequency, and the fact that 15% of
### Table 1. Trials Assessing Glargine Mixing

<table>
<thead>
<tr>
<th>Trial characteristics</th>
<th>Cengiz et al., 2010&lt;sup&gt;5&lt;/sup&gt;</th>
<th>Kaplan et al., 2004&lt;sup&gt;6&lt;/sup&gt;</th>
<th>Fiallo-Scharer et al., 2006&lt;sup&gt;7&lt;/sup&gt;</th>
<th>Hassan et al., 2008&lt;sup&gt;8&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Design</strong></td>
<td>Randomized, open-label, 2-day duration</td>
<td>Randomized, crossover, 30-day duration</td>
<td>Prospective, case-control, 12-month duration</td>
<td>Randomized, controlled, 3-month duration</td>
</tr>
<tr>
<td><strong>Regimen(s)</strong></td>
<td>Lispro versus lispro mixed with glargine</td>
<td>Nonmixed glargine + lispro or aspart versus glargine mixed with lispro or aspart</td>
<td>Mixed glargine with lispro or aspart versus nonmixed glargine plus lispro or aspart twice daily</td>
<td>Mixed glargine with lispro or aspart versus mixed NPH with lispro or aspart twice daily</td>
</tr>
<tr>
<td><strong>n</strong></td>
<td>11</td>
<td>14</td>
<td>110 (55 in each group)</td>
<td>42 (21 in each group)</td>
</tr>
</tbody>
</table>
| **Participants**      | • Type 1 diabetes  
• Ethnicity not reported  
• Mean age 15.1 ± 3 years  
• Sex not reported  
• Mean duration of diabetes not reported  
• Baseline A1C 7.6 ± 0.6% | • Type 1 diabetes  
• Ethnicity not reported  
• Mean age 13.5 ± 0.5 years  
• 6 male  
• Mean duration of diabetes 44 ± 8 months  
• Baseline A1C 7.7 ± 0.2% | • Type 1 diabetes  
• Ethnicity not reported  
• Mixed group:  
  • Mean age 13.4 ± 3.8 years  
  • 35 male  
  • Mean duration of diabetes 85 ± 45.5 months  
  • Baseline A1C 8.54 ± 1.14%  
• Nonmixed group:  
  • Mean age 12.9 ± 4.0 years  
  • 35 male  
  • Mean duration of diabetes 84.9 ± 45.1 months  
  • Baseline A1C 8.61 ± 1.14%  
• All patients using glargine and an RAI for at least 3 months before enrollment  
• 41 in mixed group were taking NPH before breakfast to cover lunch  
• 40 in nonmixed group were taking NPH before breakfast to cover lunch | • Type 1 diabetes  
• Ethnicity: 74% Caucasian (n = 31)  
• Mixed glargine group:  
  • Mean age 11.8 ± 3.7 years  
  • 10 male  
  • Mean duration of diabetes 3 months  
  • Baseline A1C 6.8 ± 1.0%  
• Mixed NPH group:  
  • Mean age 10.1 ± 2.0 years  
  • 8 male  
  • Mean duration of diabetes 3 months  
  • Baseline A1C 6.9 ± 1.0%  
• Baseline insulin regimens were not reported for either group |
| **Primary outcome**   | Maximum insulin action (peak effects) expressed as glucose infusion rates (mg/kg/min) lispro separate 7.1 ± 1.8  
Maximum glucagon response 2.5 ± 0.6 | Glucose levels (using continuous glucose monitoring in 20-min intervals for 24 h):  
NSD in preprandial (158.4 ± 162, 149.4) versus NPH (306, 163.8)  
Maximum concentration mlU/ml):  
lispro separate 149 ± 29%  
Maximum concentration mlU/ml):  
lispro separate 55 ± 19 min  
Fasting blood glucose at 3 months: glargine RAI 102.6 ± 342 versus NPH + RAI 91.8 mg/dl, P = 0.008  
Hypoglycemia at 3 months: glargine RAI none versus lispro mixed 3 patients (1, 2, 0.6180) and 6 months (mixed 1 vs. nonmixed 0, P = 0.4731)  
NSD in hypoglycemia: 5% baseline, 9% separate injections  
NSD in severe hypoglycemia at 3 months (mixed 3 vs. nonmixed 0, P < 0.027)  
NSD in hypoglycemia: 5% baseline, 9% separate injections  
NSD in severe hypoglycemia at 3 months (mixed 3 vs. nonmixed 0, P = 0.4731) | A1C at 6 months:  
nomixed 8.54 ± 1.14 versus mixed 8.61 ± 1.14%, P = 1.0  
A1C at 3 months:  
glargine + RAI 6.7 ± 1.3 versus NPH + RAI 7.6 ± 1.0%, P < 0.027 |
Table 1. Trials Assessing Glargine Mixing

<table>
<thead>
<tr>
<th>Design</th>
<th>Regimen(s)</th>
<th>Participants</th>
<th>Sex</th>
<th>Mean duration of diabetes</th>
<th>Baseline A1C</th>
<th>Baseline insulin regimens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prospective, case-control, 12-month duration</td>
<td>Nonmixed glargine + lispro or aspart versus glargine mixed with lispro or aspart</td>
<td>Type 1 diabetes</td>
<td>Ethnicity not reported</td>
<td>44 ± 8 months</td>
<td>7.6 ± 0.6%</td>
<td>Not reported</td>
</tr>
</tbody>
</table>
| Randomized, crossover, 30-day duration | Mixed group: Mean age 15.1 ± 6 years | Male | 45.5 months | 1.14% ± 1.0% | A1C at 3 months: glargine RAI 6.7 ± 1.3% versus lispro separate 7.1 ± 1.0% \(P = 0.04\) | \(n = 17\) lost to follow-up:
- Mixed: 11 (4 because of increased incidences of hypo- and hyperglycemia and 7 who failed to follow up within 3 months after enrollment)
- Nonmixed: 6 (all failed to follow up within 3 months after enrollment)

| Notable secondary outcomes | Maximum concentration mlU/ml: lispro separate 149 ± 38 versus lispro mixed 53.9 ± 9 | Time to maximum concentration: lispro separate 55 ± 6 min versus lispro mixed 106 ± 19 min | A1C at 3 months: NSD, \(P = 1.0\); actual values not reported | Fasting blood glucose at 3 months: glargine + RAI 102.6 ± 36 versus NPH + RAI 172.8 ± 91.8 mg/dl, \(P < 0.008\) Lunch and supper blood glucose at 3 months: NSD |
| Safety outcome(s) | None reported | NSD in hypoglycemia: 5% baseline, 9% separate injections, and 2% mixed injections; \(P\) not reported | NSD in nonsevere hypoglycemia at 6 months \((P = 0.6134)\); incidence not reported NSD in severe hypoglycemia at 3 months (mixed 3 vs. nonmixed 1, \(P = 0.6180\)) and 6 months (mixed 1 vs. nonmixed 0, \(P = 0.4731\)) NSD in diabetic ketoacidosis at 3 months (mixed 1 vs. nonmixed 2, \(P = 1.0\)) and 6 months (mixed 1 vs. nonmixed 0, \(P = 0.4731\)) | Hypoglycemia at 3 months: glargine + RAI none versus NPH + RAI 3 patients (1, 2 and 4 times each), NSD |
| Withdrawals | None | 9% \((n = 1)\) excluded because of noncompliance with study regimens | 15% \((n = 17)\) lost to follow-up: mixed: 11 (4 because of increased incidences of hypo- and hyperglycemia and 7 who failed to follow up within 3 months after enrollment) nonmixed: 6 (all failed to follow up within 3 months after enrollment) | 29% \((n = 12)\) lost to follow-up:
- Glargine + RAI: 8 (4 because of lost contact, 2 who failed to keep clinic visits, 1 who did not attend class, and 1 who rescinded consent)
- NPH + RAI: 4 (2 because of lost contact, 1 who failed to keep study visit, and 1 who moved away after turning 18) |

NSD, no significant difference; RAI, rapid-acting insulin (lispro or aspart)
patients were lost to follow-up in the first 3 months (n = 11 in the mixed group and n = 6 in the nonmixed group).

Hassan et al.\(^8\) conducted a 12-week randomized, controlled trial in 42 pediatric patients (18 males; mean age 11.8 ± 3.7 years in the mixed glargine group and 10.1 ± 2.0 years in the mixed NPH group) who were newly diagnosed (<3 months) with type 1 diabetes. This trial compared the efficacy and safety of administering glargine mixed with either aspart or lispro to that of administering NPH insulin mixed with either aspart or lispro. Patients who were randomized to the mixed glargine regimen (n = 21) were converted unit for unit from their NPH dose. These patients also went to a class to learn how to mix glargine and how to calculate doses of their RAI based on carbohydrate counting and their insulin sensitivity factor. Patients who were randomized to the mixed NPH regimen (n = 21) were continued on their previous dose at the time of enrollment and did not attend an educational class. The primary outcome was change in A1C.

There was a significant difference in A1C levels at 3 months with mixed glargine compared to mixed NPH (6.7 ± 1.3 vs. 7.6 ± 1.0%, \(P < 0.027\)). There also was a significant difference in fasting blood glucose levels (\(P < 0.008\)) with mixed glargine compared to mixed NPH (Table 1). There was no significant difference in the occurrence of hypoglycemia between the treatment groups (Table 1).

Strengths of this trial included its prospective, randomized design and intention-to-treat analysis. Limitations included a lack of standardized dosing, lack of blinding, 15% dropout rate, and lack of standardized, equal patient contact. Another limitation is the comparison of NPH to glargine. A few trials\(^9\)–\(^11\) conducted in adolescents with type 1 diabetes have reported significant lowering in A1C (ranging from −0.3 to −1.0%) when transitioned from NPH to glargine, whereas others\(^12\),\(^13\) have reported no significant change in A1C values. None of these studies\(^9\)–\(^13\) mixed glargine with an RAI.

In the trial by Hassan et al.\(^8\), patients maintained similar A1C levels in the glargine group (baseline 6.8 ± 1.0% and 3 months 6.7 ± 1.3%) when converted from NPH. Blood glucose control declined when patients were maintained on NPH in the mixed NPH group (A1C at baseline 6.9 ± 1.0% and 3 months 7.6 ± 1.0%). The authors did not offer an opinion as to reasons for loss of control in the mixed NPH group.

Summary
Of the four studies assessing mixing of insulin, only one was a pharmacokinetic study assessing peak effects of insulin action, maximum insulin concentrations, and time to peak concentrations.\(^3\) The other three studies\(^6\)–\(^8\) assessed clinical outcomes in real-world settings. All of these studies were conducted in pediatric patients with type 1 diabetes.

The pharmacokinetic study by Cengiz et al.\(^4\) reported significant changes in peak effects of insulin action, maximum insulin concentrations, and time to peak concentrations for lispro when mixed with glargine. However, two of the clinical outcomes studies\(^6\),\(^7\) did not report significant differences in A1C levels at 3 or 6 months or preprandial, postprandial, or nocturnal blood glucose levels when mixing glargine with an RAI. One of the clinical outcome studies\(^4\) reported improved A1C and fasting blood glucose at 3 months with an RAI mixed with glargine compared to an RAI mixed with NPH insulin. There were no significant differences in hypoglycemia in any of the clinical outcome trials at any time measured.\(^6\)–\(^8\)

There are several limitations that should be noted overall in these studies. The study population was exclusively pediatric patients with type 1 diabetes, which limits application of these results to adult patients with type 1 diabetes or to patients with type 2 diabetes. The study populations were very small, with the largest including 110 enrolled patients. Larger studies would help to validate or refute these initial findings and allow application to a broader patient population. The study durations were also quite short (the longest “mixed” period being 6 months), and longer study durations are needed to ensure that safety and efficacy can be adequately assessed.

Conclusion
Limited clinical trials have demonstrated no significant changes in clinical outcomes (blood glucose levels, A1C levels, and hypoglycemia) when mixing glargine with an RAI. However, these trials were small and included only pediatric patients. Additional studies with larger patient populations and longer trial durations are needed before mixing glargine with an RAI can be recommended on a routine basis in clinical practice.

The educational process needs to be an unending process involving patients, families, caregivers, and the health care team. Many factors, including needle length and injection technique, can contribute to pain from insulin injections. Demonstration of appropriate insulin delivery and site rotation during office visits can help avoid complications related to the use of insulin. Health care providers should also regularly examine injection sites for bruising, infections, lipoatrophy, or lipohypertrophy.

Patients may be independently mixing their glargine with their RAI to minimize the number of daily injections. Therefore, insulin dosing and delivery should be addressed regularly with all patients. The routine mixing of glargine and RAIs should not be recommended until additional data are available to support this practice.

References

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