Diabetes is associated with microvascular and macrovascular complications leading to significant morbidity and mortality. Glycaemic control is important to prevent and delay the progression of these complications. An ideal insulin regimen in patients with diabetes would mirror the 24-hour insulin profile of a non-diabetic person and thereby prevent hyperglycaemia without inducing hypoglycaemia. This has, until recently, proved difficult to reproduce by regular subcutaneous insulin injections due to the inherent pharmacokinetic properties of the available insulins. Normoglycaemia was rarely achieved without hypoglycaemia compromising the quality of patients’ lives. The advent of the new long- and short-acting insulin analogues are expected to both improve glycaemic control leading to a reduction in diabetes-related complications and reduce the incidence of hypoglycaemia thereby offering patients a better lifestyle.

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**Key words:** insulin, pharmacology, analogues, lispro, aspart, glargine, detemir.

**Introduction**

Insulin, from the pancreatic beta-cells, plays a crucial role in glucose homeostasis. It acts on the liver to inhibit glycogenolysis and gluconeogenesis, and in the periphery to stimulate glucose utilisation by muscle and fat. An ideal insulin treatment regimen for people with diabetes would reinstate the normal daily insulin profile (figure 1) and prevent hyperglycaemia without inducing hypoglycaemia.

However, in contrast to the physiological delivery of insulin into the hepatic portal circulation, subcutaneously injected insulin provides relatively less insulin to the liver than the periphery. Hence, the normal balance of hepatic and peripheral effects of insulin cannot be exactly restored by conventional injections.

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**Figure 1.** The normal daily profile of plasma insulin

Adapted with permission from Day et al. *Br J Cardiol* 2003;10:379-83.
at positions A8 and A10 (respectively threonine and isoleucine in man versus alanine and valine in the ox). Animal and human insulin are therefore nearly homologous and the differences are not crucial to the receptor binding or action of insulin.

The carboxyl end of the B chain of the insulin molecule is involved in self-association of two insulin molecules into dimers. Therefore, the self-association properties of human and porcine insulin may be different.1 B30 threonine adds an extra hydroxyl group to the human insulin molecule, increasing its hydrophilic properties and decreasing the lipophilic properties when compared to pork insulin. Therefore, human insulin is more soluble in aqueous solutions than porcine insulins.1 The spherical structure of the insulin monomer is important for its interaction with the insulin receptor: porcine insulin has a slightly different three-dimensional structure to human insulin.2

However, there is no measurable difference in biological potency of human and animal insulins.3 Human insulin is absorbed slightly faster from a subcutaneous injection site1 than porcine insulin, which may be explained by greater hydrophilicity of the human insulin molecule.2

Short-acting insulins

Standard

Insulin in neutral concentrated solutions in vials is mostly zinc-containing hexamers. For absorption to occur after subcutaneous injection, insulin needs to be in a monomeric form. As dissociation of the insulin hexamers to monomers is slow, so is the rate of absorption from the subcutaneous site.4 For example, it can take approximately 1–2 hours for absorption of subcutaneous regular short-acting insulin.1 This results in a slow rise to peak insulin concentrations and hence suboptimal plasma insulin levels in the early phase of glucose absorption after a meal, leading to postprandial hyperglycaemia. The insulin levels also fall slowly after the peak, extending the period of elevated insulin concentrations with the risk of later hypoglycaemia.6 The influence of short-acting insulins on post-prandial hyperglycaemia is improved by subcutaneous administration 30 minutes to one hour before eating6 but the delayed risk of hypoglycaemia remains. Therefore, the extent of the insulin crystallisation and the ease of dissociation play an important role in determining the rate of absorption after subcutaneous injection.

Rapid-acting analogues

Insulin hexamer formation is an obstacle to mimicking physiological insulin profiles after subcutaneous injection. This led to the hypothesis that a reduced tendency of insulin to self-associate would enable faster absorption and shorter duration of action. Using this rationale monomeric insulin analogues were created by alteration of selected amino acid residues in regions vital for self-association.7 After subcutaneous injecting they are absorbed 2–3 times faster than the regular non-dissociating hexameric insulins7,8 giving a prompt rise in circulating insulin and the opportunity to inject just before a meal. The rates of disappearance are also 2–3 times faster with no lag phase,8 so monomeric insulins result in a more physiological prandial plasma insulin profile.

The two currently available rapid-acting insulin analogues are insulin lispro (LysB28, ProB29) and insulin aspart (AspB28) (figure 2).

Insulin lispro (LysB28, ProB29-human insulin)

Insulin lispro, the first insulin analogue to be used clinically, has the B28 (proline) and B29 (lysine) residues inverted, resulting in reduced self-association.7,9 This inverted sequence was found and thought to be responsible for the lower degree of self-association of insulin-like growth factor (IGF-1) and insulin lispro was engineered to incorporate this pharmacological advantage.10 Both human insulin and insulin lispro exist in their respective formulations as hexamers that are stabilised with zinc and phenolic preservatives to assure two years of shelf life at 4°C. However, insulin lispro differs in that its hexamer complex rapidly dissociates.
into monomeric subunits, as the phenolic ligands diffuse away in the interstitial space resulting in a plasma absorption profile indistinguishable from that of pure monomeric insulin.4,11

Immunogenicity, receptor binding and metabolic effects of insulin lispro are similar to human insulin.12-14 Lispro does however have a slightly elevated IGF-1 receptor affinity.14 Toxicology studies, which included reproductive and developmental periods in animals, showed no long-term toxicity4 and pooled data from clinical studies showed no increase in adverse events or progression of diabetic complications.15

The rapid absorption, onset and short duration of action of lispro results in a more precise insulin profile at mealtimes with a peak approximately one hour after subcutaneous injection and rapid elimination. After intravenous injection ‘regular’ human insulin and insulin lispro have identical pharmacokinetics and pharmacodynamics.12,16

Glycaemic control on insulin lispro is comparable17 or improved18,19 compared to ‘regular’ short-acting insulin. Improved glycaemic control appears to be best achieved with multiple injection regimens where the basal dose is optimised,20,21 although this can be associated with increased hypoglycaemia.22 Insulin lispro can be mixed with neutral protamine Hagedorn (NPH) without any pharmacokinetic changes.21 Several studies have also shown improved post-prandial glucose levels in type 117,24,25 and type 2 diabetes26 including adolescents.27 This improvement was at the expense of increased fasting and preprandial glucose levels in one study,28 although fasting glycaemic control was not significantly different in a meta-analysis.17

Insulin lispro reduced hypoglycaemia in several studies compared to regular human insulin, especially at night-time.21,24,28 Severe hypoglycaemia was also reduced, possibly due to purportedly enhanced hepatic sensitivity to glucagon with insulin lispro.29 However, the counterregulatory hormone responses after insulin lispro compared to human insulin and porcine insulin were similar during a stepwise euglycaemic/hypoglycaemic clamp.30 Further studies showed the counterregulatory hormone responses on insulin lispro were improved on relatively conserved and equivalent to physiological responses to hypoglycaemia in patients on regular insulin.31

Although the optimal time for injecting insulin lispro is immediately before the meal, it can be injected up to 15 minutes after a meal.30 Treatment satisfaction and flexibility scores on insulin lispro were improved compared to regular human insulin.31 Insulin lispro appears to be as safe as regular human insulin in pregnant women with type 1, type 2 and gestational diabetes.32-34 It does not cross the placenta after a single standard dose35 and does not cause adverse outcomes in pregnancy.36 Concerns about progression of retinopathy have not been demonstrated,37 but further prospective trials are required to confirm these findings.

Insulin aspart (Novorapid)
Insulin aspart is formed by replacing the proline at B28 with a negatively charged aspartic acid residue. This switch results in reduced self-association of the insulin molecule. Insulin aspart initially exists as hexamers, which after subcutaneous injection rapidly dissociate into dimers and monomers.4 Aspart resembles human insulin in receptor binding, potency38 and metabolic and mitogenic effects.39 No safety issues were raised by a range of experiments in which insulin aspart and human insulin exhibited similar pharmacological profiles.40 It was associated with an increase in cross-reactive insulin antibodies, which subsequently fell towards baseline values, without any apparent effects on efficacy or safety.40

After subcutaneous administration insulin aspart is absorbed faster than ‘regular’ human insulin, with higher peak insulin concentrations and a reduced duration of action.41,42 Aspart improves post-prandial glucose control43 and in some studies it improves glycaemic control44,45 compared to regular insulin in patients with type 1 diabetes. It has been associated with reduced night-time and severe hypoglycaemia in some45 but not all studies.46 The counterregulatory and symptomatic responses to acute hypoglycaemia between patients on insulin aspart and regular human insulin are similar.47 Aspart can be administered immediately prior to a meal or postprandially48 and treatment satisfaction and quality of life scores were significantly improved compared to ‘regular’ human insulin.49 No adverse effects were seen with insulin aspart46 but it is of note that no safety data in pregnancy are available.

It is likely that insulin aspart, like insulin lispro, will require multiple injection regimens where the basal dose is optimised. Insulin aspart can be mixed with NPH without alteration of its pharmacokinetics.49

Long-acting insulins
The first successful insulin preparation with a prolonged action was protamine insulin followed by the Lente crystalline zinc-insulin series. Classically long-acting insulins have been formulated as crystalline or amorphous suspensions obtained by addition of small amounts of zinc that at high insulin concentrations and neutral pH increase the self-association into dimers and hexamers. The resulting insulin is absorbed slowly from the injected depot. The current long-acting insulins are Ultralente (Ultratard), the longest acting human insulin-zinc crystalline suspension, NPH (or isophane) insulin, an intermediate acting insulin protamine suspension and Lente (70/30 mixture of ultralente with semilente), an intermediate acting insulin (table 1).49

<table>
<thead>
<tr>
<th>Human insulin type</th>
<th>Onset of action</th>
<th>Peak of action</th>
<th>Duration of action</th>
</tr>
</thead>
<tbody>
<tr>
<td>Regular</td>
<td>30–60 minutes</td>
<td>2–4 hours</td>
<td>6–8 hours</td>
</tr>
<tr>
<td>NPH</td>
<td>1–3 hours</td>
<td>5–7 hours</td>
<td>13–18 hours</td>
</tr>
<tr>
<td>Lente</td>
<td>1–3 hours</td>
<td>4–8 hours</td>
<td>13–20 hours</td>
</tr>
<tr>
<td>Ultralente</td>
<td>2–4 hours</td>
<td>8–14 hours</td>
<td>18–30 hours</td>
</tr>
</tbody>
</table>

Regular human insulin is shown for comparison. Adapted from Hirsch I. Med Clin N Am 1998;82:689-719.
Key messages

- Physical and chemical properties of insulin have been used to alter subcutaneous absorption
- Recombinant DNA technology has led to the production of novel insulin analogues
- The new short-acting insulin analogues allow more flexibility with rapid absorption, onset and short duration of action
- The new long-acting insulin analogues allow a more physiological basal insulin profile

Crystalline insulin suspensions have a number of disadvantages. Ultratard, although it has a long duration of action, has a high intersubject and intraindividual variation of absorption. This is partly due to suboptimal mixing of the insulin suspension. NPH and Lente are usually too short-acting for a once-a-day dosing. NPH peaks in action 5–7 hours after injection and so the night-time dose can cause nocturnal hypoglycaemia in the early hours when insulin sensitivity is often increased. Reducing the evening dose, however, results in raised morning blood glucose due to a relative reduction in insulin sensitivity between 0500–0800 hours (known as the dawn phenomenon).50

Long-acting analogues

To achieve physiological insulin profiles ideal basal insulins should have a 24-hour peakless activity profile allowing once-daily dosage, with little variability in blood glucose excursions.50

Two approaches are being used to develop new long-acting insulin analogues.

The first approach changes the isoelectric point (the pH at which insulin is least soluble and precipitates) resulting in an insulin molecule that is soluble at body pH (and hence eliminates the variability problems with suspensions) but precipitates in subcutaneous tissues where the pH is near neutral.

Insulin glargine (A21 Gly, B31 Arg, B32 Arg) was developed using this method (figure 2). It is a long-acting human insulin with reduced incidence of hypoglycaemia.51 The addition of two analogue that has a near peakless action for at least 24 hours using this method (figure 2). It is a long-acting human insulin that states in subcutaneous tissues where the pH is near neutral.

The second approach involves the addition of a fatty acyl chain, which allows the insulin to bind to non-esterified binding sites on albumin. As the modified insulin dissociates slowly from albumin and favours the hexameric state the resultant absorption and duration of action are longer. This is the rationale behind the current development of insulin detemir.49

Insulin detemir is a soluble long-acting analogue with a fatty acyl chain attached to B29 Lys (figure 2). In healthy subjects detemir has a less pronounced peak compared to NPH insulin. Maximum concentrations were reached after 4–6 hours,46 and detemir appears to produce a more predictable glycaemic control, a smoother plasma glucose profile with a significant reduction in hypoglycaemia. Further human pharmacodynamic and clinical studies are awaited.

Conclusions

New long- and short-acting insulin analogues are providing greater opportunity for patients to improve glycaemic control. This should lead to a reduction in diabetes-related complications and reduce the incidence of hypoglycaemia, thereby offering patients a better lifestyle.

References