1. Galegine and antidiabetic plants

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The modern pharmacopoeia contains many examples of folk remedies which have led to the discovery of important therapies for a range of conditions. Early experiments with extracts of the herb, *Galega officinalis*, led to the characterization of the blood glucose lowering effects of galegine. The discovery of metformin, today's foundation therapy for type 2 diabetes, can be traced back to this early pioneering work.

Pharmacognosy of diabetes mellitus

The principal clinical features of diabetes mellitus were recognized as long ago as about 1500 BC, when Hindu scholars described a condition featuring polydipsia, polyuria, wasting away of the body and the production of urine sweet enough to attract flies and ants [1]. The Ebers Papyrus, held in the University Library, Leipzig, Germany, shows that diabetes was also recognized in ancient Egypt, and recommends a diet of fruits, grains and honey for those afflicted. Pharmacognosy (the study of the medicinal properties of materials of natural origin) has played an important role in the management of diabetes mellitus since that time.

Indeed, it has been estimated that more than 400 herbal or plant-derived products have been used for the management of type 2 diabetes across geographically and culturally diverse populations worldwide [2]. These preparations, often derived from ancient use of folk medicines, include garlic, onion, ginseng, bitter melon, fenugreek, *Gymnema sylvestre*, *Pterocarpus marsupium* and other plants containing the flavonoid compound, epicatechin, bilberry, aloe vera, and holly [3]. A further plant-derived substance with antidiabetic properties, galegine, is discussed in detail, below. Interest in this approach shows no sign of abating; a search of the Medline database reveals evaluations of plant-derived antidiabetic medicines by indigenous North American tribes [4], and in India [5], Central America [6,7], Europe [8], and North Africa [9] within the last 15 years.

The active ingredients of a number of plant-derived antidiabetic preparations have been identified, and potentially beneficial effects on the rate of food ingestion, glucose transport, potentiation of insulin release, inhibition of insulin clearance, insulin-mimetic effects, reduced gluconeogenesis, and β -cell protection have been attributed to these agents [3]. However, a recent systematic review of plant-derived antidiabetic agents has found little evidence for efficacy in controlling blood glucose, although several were identified as deserving further study. Nevertheless, such evaluations are important. Purified constituents of natural (plant or animal) products have provided important therapeutic agents that have achieved widespread use in several areas of medicine. Alternatively, a natural product often provides a research tool that facilitates our understanding of key elements of physiology.

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Metformin: the gold standard

Galegine, a substance produced by the herb *Galega officinalis*, provides an excellent example of such a discovery. Experimental and clinical evaluations of galegine, described below, provided the pharmacological and chemical basis for the subsequent discovery of metformin, today's foundation therapy for type 2 diabetes uncontrolled by diet and exercise therapy.

Galega officinalis: from botany to bedside *Overview of Galega officinalis*

Galega officinalis (Figure 1.1) is a summer-flowering hardy perennial that originated in southern Europe and western Asia, but has now spread to many other countries around the world, where it is known under a number of other names, including Goat's Rue, Spanish sanfoin, false indigo, Italian fitch, French lilac, and Professor-weed. In some cases, *G. officinalis* was introduced deliberately. For example, *G. officinalis* has long been cultivated as a garden plant, and has been used for diverse medicinal purposes since medieval times ("*officinalis*" signifies "sold as a herb"), including for control of the polyuria associated with diabetes and for promotion of perspiration in people with the plague [10,11].

In addition, *G. officinalis* is believed to have been introduced into the USA in the 1800s as a potential forage plant [12]. It had been observed that its ingestion could stimulate lactation in livestock and the name, "Galega", is believed to derive from the Greek terms for milk (gala) and goat (aigos) [11]. Indeed, research into this effect of *G. officinalis* continues today [13]. Unfortunately, the plant has proved too toxic for widespread agricultural use, with the potential to induce tracheal frothing, pulmonary oedema, hydrothorax, hypotension, paralysis and death [11,14] and it





Galegine



Metformin



Figure 1.2 Chemical structure of galegine in comparison with that of metformin.

is widely considered to represent a threat to grazing animals. For example, many states of the USA now classify *G. officinalis* as a noxious weed, with considerable effort and expense committed to its eradication [15].

Initial preclinical evaluation of galegine

The therapeutic potential of *G. officinalis* for the management of diabetes was defined in the first half of the twentieth century. *G. officinalis* is a rich source of guanidine and related molecules, which account for its biological effects. The toxicity of guanidine precludes its use clinically, and experiments by Georges Tanret in the years immediately before the Great War identified a less toxic guanidine-like alkaloid, galegine. The precise structure of galegine was confirmed as isoamylene-guanidine (Figure 1.2) by a group in Edinburgh, UK, in 1923 [16]. The contribution of researchers at Edinburgh University was described in a review by Hadden [17]. This reproduced plates from "The Vegetable System" [18], a book published in 256 volumes in the latter half of the eighteenth century, now resident in the library of the Royal College of Physicians of Edinburgh. These plates are reproduced here as Figure 1.3.

Tanret returned to the study of galegine after the war, and published a preclinical account of the pharmacology of this agent in rabbits and dogs in 1927 [19]. In fact, this study resembles a modern toxicology study in many respects, as it involved the administration of increasing doses of galegine until symptoms reminiscent of hypoglycaemia were encountered. The dose-response relationship of this agent was steep. In rabbits, subcutaneous injections of galegine at an average dose of 150 mg/kg induced little effect on blood glucose. A slight increase in the dose to about 300 mg/kg induced a mild transient lowering of blood glucose in some animals, while further dose increases to an average of about 380 mg/kg or more induced an increasing frequency of hypoglycaemic crises and death in the hours following injection. Oral administration at doses at the top of this range induced a similar reaction (Figure 1.4). Blood glucose responses were variable, and most animals did not develop a hypoglycaemic response. However, some rabbits developed profound hypoglycaemia soon after oral administration of galegine, leading to death in some cases. Dogs were more sensitive to galegine-related toxicity. Interestingly, the hypoglycaemic actions of galegine sulphate were also present in pancreatectomised animals.

Metformin: the gold standard



Figure 1.3

Reproductions of plates describing *Galega officinalis* from "The Vegetable System", published in the second half of the eighteenth century [17,18].



Figure 1.4

Effect of oral administration of galegine sulphate on blood glucose in normoglycaemic rabbits.

Asterisks indicate death of animals from hypoglycaemia. Glycaemia is shown in original units. One part per thousand of glucose equates to 5.6 mmol/L. Doses were given as fixed numbers of centigrams of galegine sulphate per kg; as weights of animals were also provided, this permitted calculation of doses in mg/kg. Blood glucose values have been converted from the original units used (g/L). Drawn from data presented by Simmonet and Tanret [19].

In some of the experiments of Simonnet and Tanret, hyperglycaemia preceded or followed a hypoglycaemic response (visible in one animal shown in Figure 1.4). Müller and Reinwein also described the preclinical pharmacology of galegine sulphate and also encountered this phenomenon at higher doses [20]. At a dose of 4 mg/kg in dogs, galegine sulphate induced a transient hyperglycaemia in some animals, but this was followed by reductions in blood sugar of 40% or more typically occurring over 11–12 hours after dosing.

Effects of galegine in humans

Müller and Reinwein also described early clinical experience with galegine sulphate [20]. An initial experiment involved self-administration of a dose of 109 mg galegine sulphate, after which they followed blood glucose levels for 25 hours. Following further dose-ranging experiments in healthy individuals, these studies were then extended to patients with diabetes. Figure 1.5 shows the effects of galegine on blood glucose levels in all three subjects, in the case of the diabetic subjects with or without a prior high-fat meal. Initial blood glucose was higher in the diabetic patients, as would be expected. In all three subjects, a hypoglycaemic effect was observed (mild in the normoglycaemic subject but marked in the diabetes patients), after which blood glucose levels returned to the values close to pre-treatment level.

Further work by Leclerc [21] and by Parturier and Hugonot [22] during the following decade added further observations on the antidiabetic actions of extracts of *G. officinalis*. These sought to improve the delivery and safety of galegine-based therapy, and may have succeeded to some extent. However, the variability in responses to the treatment, and its short duration of action limited their utility [11].

Other early clinical studies on guanidine derivatives

In addition, numerous other guanidine derivatives were studied around this time, including a series of biguanide derivatives [23,24]. Two of these, decamethylene diaguanide (known as Synthalin A) and dodecamethylene diaguanide (Synthalin B) did achieve some clinical use [25].

A further agent, butylamin-guanidin, also described as "synthalin" was evaluated in humans. A report from Montreal in 1927 describes mixed success with this agent, with some patients "sugar



Figure 1.5

Effects of galegine sulphate on blood glucose in humans.

Blood glucose values have been converted from the original units used (grams per cent). Drawn from data presented by Müller and Reinwein [20].

Metformin: the gold standard

free", some able to reduce their insulin dose, and some not showing any clinical response [26]. Side-effects included abdominal pain and cramping, and nausea. Interestingly, this report describes lower body weights with the guanidine derivative than with insulin, a benefit well described today for metformin [27]. Elliott P Joslin evaluated synthalin, and described his results (similar to those described above) in a brief report to the Nineteenth Annual Meeting of the American Society for Clinical Investigation in 1927 [28].

Significance of these early studies

The early experiments described above clearly lacked the rigour and structure of today's highlyregulated clinical research and the increasing availability of insulin as an antidiabetic therapy diverted attention from medicinal chemistry of prototype oral antidiabetic agents. As Joslin said, in his report to the American Society for Clinical Investigation in 1927 [28]:

The extraordinary improvement of the modern diabetic with diet and insulin makes it exceedingly difficult to estimate the value of any new method of treatment.

Nevertheless, they represent the first attempts to properly characterize oral pharmacotherapy for diabetes built on an evidence base of published material. The following chapters of this book show how this pioneering work led to the discovery of metformin, still the foundation therapy for type 2 diabetes at the beginning of the twenty first century. Again, Joslin's own words [28], referring to his experiments on synthalin, ring true today and form an eloquent postscript to this pioneering work:

More important is the probability that it is the forerunner of other and better preparations which one can give by mouth and which to a certain degree will replace insulin.

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