ABSTRACT: Rational treatment of diabetic polyneuropathy depends upon establishing its cause, which is at present unknown. A number of animal models of diabetes have been examined and although abnormalities are detectable in the peripheral nervous system they do not duplicate the degenerative neuropathy encountered in the human. The relevance of these abnormalities is therefore uncertain, although they may reflect the earlier changes in man. For human neuropathy, it is likely that vascular lesions or an abnormal susceptibility to mechanical injury are responsible for focal neuropathies. The evidence that ischaemia and hypoxia are responsible for the diffuse sensory neuropathy and autonomic polyneuropathy is still equivocal and it is often difficult to establish whether the vascular changes are primary or secondary. Metabolic explanations, such as sorbitol accumulation in nerve, have not so far been adequately validated by responses to treatment. The manifestations of diabetic neuropathy are complex and a single explanation should not be sought.

Satisfactory treatment for diabetic neuropathy is currently not available. It is known that strict glycemic control, either by continuous subcutaneous insulin infusion or pancreatic transplantation will stabilize the neuropathy, although little improvement has been detected over periods of up to 2 years. Neither of these forms of treatment is practical except in special circumstances or for research purposes. With conventional treatment it is difficult to obtain satisfactory glycemic control in a high proportion of patients, either because of poor compliance or because of ‘brittle’ diabetes. It is therefore imperative to establish the mechanism of diabetic neuropathy so that, if possible, its occurrence can be prevented even if adequate glycemic control cannot be obtained. This article will review current ideas as to the causation of peripheral neuropathy in patients with diabetes mellitus, but first it is necessary to describe briefly what is being caused. It is clear that a wide variety of disturbances of peripheral nerve function may be encountered.

CLASSIFICATION OF DIABETIC NEUROPATHY

It is convenient to recognize two broad categories of peripheral nerve disturbance related to diabetes. These can be termed rapidly reversible phenomena and more persistent or ‘established neuropathy’.

Rapidly reversible phenomena include evanescent sensory symptoms such as pain and dysesthesia, experienced mainly in the lower limbs in poorly controlled patients, that rapidly resolve once glycemic control is established. The second is the

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Received September 10, 1991. Accepted October 8, 1991
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reduced nerve conduction velocity that is found in such patients and which improves with a time course that is too rapid for remyelination or regeneration to have been responsible. Finally, there is the phenomenon of abnormal resistance to ischemic conduction failure, shared by other groups of patients such as those with uremia, hepatic failure, chronic respiratory failure and motor neuron disease, and which is also seen with aging. At present there is no indication that these rapidly reversible phenomena are predictors for established diabetic neuropathy. They will therefore not be considered further.

More persistent or 'established' diabetic neuropathy is again subdivisible into two categories. The first consists of focal and multifocal neuropathies involving isolated cranial nerve palsies, thoracoabdominal neuropathy, focal limb neuropathies and the proximal asymmetric lower limb motor neuropathy (diabetic amyotrophy). The second category includes approximately symmetric syndromes of which the most important examples are the sensory and autonomic polyneuropathies. These constitute the commonest types of diabetic neuropathy and once they have appeared are largely irreversible. They frequently coexist. The second category also includes the reversible syndrome of acute painful diabetic neuropathy and the chronic proximal symmetric lower limb motor neuropathy.

Animal Models of Diabetic Neuropathy
A number of different models of diabetes have been used in attempts to provide an acceptable animal model for human diabetic neuropathy. Those most extensively investigated include the destruction of pancreatic beta cells by alloxan or streptozotocin, and spontaneous diabetes in the BB Wistar rat, both of which are models of type 1 insulin-dependent diabetes, and the recessively inherited diabetic (C57BL/Ks,db/db) mouse, a model of type 2 non-insulin-dependent diabetes. All share impairment of the normal maturational increase in nerve conduction velocity and nerve fiber size. Loss of nerve fibers or demyelination that is any way comparable to that seen in human diabetic neuropathy does not occur, but this may be a question of the duration of the diabetes. Sima et al. have described paranodal swelling and 'axoglial junction' in the BB rat, and also in the streptozotocin-diabetic rat, but these features have not been found either in the streptozotocin-diabetic rat or in the db/db mouse in personal observations. How useful these animal models are for the interpretation of diabetic neuropathy is therefore problematic. It is hoped that they may provide clues to events occurring in the early stages of the human disease.

The Causation of Diabetic Neuropathy
Once the possible mechanisms for diabetic neuropathy are considered, the mayhem becomes evident. There has been a wealth of conflicting proposals. Currently the main contending views are those in which vascular disease is considered to underlie the totality of diabetic neuropathy and those in which a metabolic basis is favored, at least for the symmetric polyneuropathies.

The Vascular Hypothesis
Large Vessel Disease
Ischemia and hypoxia in the causation of diabetic neuropathy have had a chequered career. At the time the clinical features of diabetic neuropathy were being delineated a century ago, one of the earliest suggested causes was ischemia. This view was reiterated by Woltman and Wilder, who attributed the peripheral nerve damage to large vessel occlusive vascular disease. This study, although it had a major impact, suffered from the drawback that it was based partly on observations on nerve from amputated limbs and partly on autopsy material from patients who had had longstanding diabetes, in both of which the likelihood of coexistent atherosclerosis would have been high. Moreover, Renaut corpuscles, a normal feature of peripheral nerve, were misinterpreted as infarcts.

Peripheral nerve involvement in patients with occlusive vascular disease, although it occurs, tends not to be a major component of the clinical picture. The demand for oxygenation by peripheral nerve is presumably less than that by other tissues in the lower limbs. Chopra and Hurwitz compared the changes in nerve conduction in patients with lower limb ischemic vascular disease with those in patients with diabetic neuropathy and found distinct differences. The reduction in nerve conduction velocity was substantially greater in patients with diabetic neuropathy, this being correlated histologically with more pronounced demyelination in nerve biopsies. It has come to be accepted that large vessel occlusive disease does not play an important role in the production of peripheral nerve damage in diabetic neuropathy. Nevertheless, accompanying large vessel disease may influence the manifestations of diabetic neuropathy. The pattern of sensory loss differs in patients with diabetic polyneuropathy with and without evidence of accompanying peripheral vascular disease (P.J. Watkins, personal communication). Loss of vibration sense is more pronounced in the presence of vascular disease.

Small Vessel Disease
Interest in microvascular disease in diabetes began with the studies of Lundbaek which were applied to peripheral neuropathy by Fagerberg, who found evidence of abnormality affecting the small blood vessels in nerve biopsies from patients with diabetic neuropathy. This took the form of thickening of the walls of endoneurial vessels by PAS-positive material, later shown by electron microscopy to consist of reduplicated basal lamina. Microvascular disease was therefore proposed as the explanation for diabetic neuropathy. Other investigators, however, were unable to find a good correlation between neuropathy and small vessel disease, and possible metabolic explanations began to assume a dominant position. In recent years vascular disease has been revived as an explanation both for focal and generalized diabetic neuropathy. The evidence for and against this view will now be analyzed. In doing so it is helpful to distinguish between focal and multifocal neuropathies, on the one hand, and the distal symmetric sensory/autonomic polyneuropathy on the other.

Focal and Multifocal Neuropathy
Although not substantiated by epidemiologic studies, it is generally accepted that focal neuropathies are more frequent in patients with diabetes mellitus than in the general population. These are often at the common sites of entrapment or external compression and appear to reflect an unexplained susceptibility of diabetic nerve to compression. Whether ischemia plays any part in this is not established. In entrapment neuropathy, the...
changes in nondiabetic subjects indicate that the major pathologic feature is displacement of myelin\textsuperscript{22} although the production of the acroparesthesiae in the carpal tunnel syndrome is likely to depend upon superimposed ischemia.\textsuperscript{23} Arguments that the carpal tunnel syndrome is ischemic in origin\textsuperscript{24} are not persuasive. It is not known whether the pathology for entrapment neuropathies in diabetic individuals differs from that in nondiabetic subjects.

The acute onset of focal neuropathies, such as an isolated third cranial nerve palsy, would be consistent with an ischemic basis, although the usually prompt recovery is not typical of ischemic peripheral nerve lesions. It is explained by the underlying pathology which is demyelinating,\textsuperscript{25} this again being atypical of vascular lesions in nerve.\textsuperscript{26} In an extensive serial section study on a patient who died 3 weeks after developing an acute third nerve lesion, Asbury, et al.\textsuperscript{25} found a focal area of centri-fascicular demyelination with preservation of axonal continuity. Although not clearly related to vascular occlusion, it was considered to lie in a ‘watershed’ area and thus to be consistent with an ischemic process.

The report by Raff et al.\textsuperscript{27} on a patient with a proximal asymmetric lower limb motor neuropathy (diabetic amyotrophy) provides a less convincing demonstration. As in the study by Woltman and Wilder,\textsuperscript{12} Renault corpuscles were misconstrued as infarcts. Apart from this, the evidence for ischemic damage depended upon the demonstration of areas of ‘pallor’ affecting small cross-bridging nerve bundles between larger fascicles. The interpretation of these areas, from the histologic appearances provided, is uncertain.

It has to be concluded, therefore, that although it is highly likely that many focal nerve lesions in patients with diabetes are ischemic in origin, further documentation is required to substantiate this.

**Generalized Polyneuropathies**

**Clinical Features**

The most common type of neuropathy in diabetes is a distal symmetric polyneuropathy, the manifestations of which are predominantly sensory and autonomic. Distal motor involvement may be an accompaniment, but it is relatively minor. This pattern of involvement does not immediately suggest a vascular causation. Ischemic neuropathy related to vasculitis, whether affecting larger or smaller arterioles, apart from often being painful, affects motor function to a greater extent than sensory. It certainly does not pick out autonomic function. Larger fibers are preferentially affected and unmyelinated axons tend to be spared.\textsuperscript{28}

**Pattern of Fiber Loss**

The major evidence that generalized neuropathy has an ischemic basis has been derived from histopathologic observations on autopsied cases and on nerve biopsies. Sugiura and Dyck\textsuperscript{29} and Johnson et al.\textsuperscript{30} provided clear evidence for focal areas of fiber loss in lower limb nerves and in the two patients reported by the former authors, who had an essentially symmetric polyneuropathy, it was concluded that multifocal proximal lesions had summated to produce a diffuse distal involvement. A vascular basis was considered likely. Yet the patients in both of these studies were elderly, and coexistent vascular disease would not have been unexpected and could have given rise to focal lesions superimposed on a generalized neuropathy. Indeed, there was histologic support for this in that there was evidence of an additional sensory neuropathy with greater fiber loss in dorsal than in ventral roots. Sima et al.\textsuperscript{30} found that focal nerve fiber loss was evident in sural nerve biopsies from older patients with non-insulin-dependent diabetes mellitus (NIDDM) but not in younger patients with insulin-dependent diabetes mellitus (IDDM).

The focality of nerve fiber loss was assessed by measuring the coefficient of variation for the distribution of myelinated nerve fibers in cross sections of sural nerve biopsies by Dyck et al.\textsuperscript{31} In comparison with a control series it was found that the coefficient of variation was significantly increased, which suggested the conclusion that “in diabetes nerve fiber loss is primary and multifocal” and that this indicated a vascular causation. As the series of patients investigated by Dyck et al. was elderly and mainly had NIDDM, Llewelyn et al.\textsuperscript{32} examined sural nerve biopsies from a younger series of patients with IDDM. In comparison with biopsies from an age-matched series of biopsies from organ-donor control cases, they found a coefficient of variation for diabetic nerves virtually identical to that obtained by Dyck et al. However, when the diabetic nerves were compared with those from an age-matched series of cases with type 1 hereditary motor and sensory neuropathy (HMSN), the same increase in coefficient of variation was found. There is no indication that type 1 HMSN is a vascular disorder and such multifocality of nerve fiber loss therefore cannot be used to establish an ischemic causation for a neuropathy. The reason for this pattern of fiber loss is uncertain. In a distal length-related neuropathy it could possibly be attributed to infranastic segregation of fibers in terms of their peripheral distribution, for which there is some evidence derived from the results of intra-neural stimulation in human subjects.\textsuperscript{33}

**Morphologic Changes in Vessels**

There is no doubt that abnormalities of the small neural vessels are common in diabetes. Establishing that they are the cause of neuropathy is much more difficult. A lack of precise correlation would not exclude a causative relationship because of sampling problems. Conversely, finding a close correlation would not establish causation: both the neuropathy and the vascular changes could be independent manifestations of the diabetic state or, alternatively, the vascular changes could be secondary to the neuropathy.

Dyck et al.\textsuperscript{34} found an increased number of ‘closed’ capillaries in a series of sural nerve biopsies and found that their number was positively correlated with the severity of neuropathy as assessed by an ‘index of pathology’ that combined loss of myelinated nerve fibers with abnormalities in surviving fibers. In a later publication, Yasuda and Dyck\textsuperscript{35} reported that the luminal area of endoneurial capillaries was significantly greater in diabetic patients without neuropathy than in control subjects although not significantly different between diabetic patients with neuropathy and control subjects. Despite this, more ‘closed’ capillaries were again found in the patients with diabetic neuropathy.

Other observers have failed to support these observations. Powell, et al.\textsuperscript{36} reported that luminal diameter did not differ between diabetic patients with histologic evidence of neuro-
Intraluminal Changes

The thickening of the pericapillary basal laminal zone found by Bradley et al., in their morphometric comparison of patients with neuropathy related to IDDM, type 1 HMSN and organ donor controls, confirmed endothelial cell hyperplasia and basal laminal thickening in diabetic neuropathy but found that this was also true of type 1 HMSN. The reduplication of the basal lamina may be related to capillary hyperplasia consequent in some way on the neuropathy. This probably varies between neuropathies with different etiologies, and examination of other disorders would be of interest as it may provide an explanation. However, the occurrence of such changes in a neuropathy such as type 1 HMSN means that in themselves they cannot be used to substantiate a vascular causation for diabetic neuropathy.

The thickening of the pericapillary basal laminal zone found by Bradley et al. was greater in the patients with diabetic neuropathy than in those with HMSN, although the difference did not reach statistical significance. Nevertheless, there were interesting differences between the diabetic and type 1 HMSN patients. In the former the layers tended to remain intact whereas in the latter they became fragmented. This is possibly related to increased resistance of the basal laminal material to proteolytic enzymes because of nonenzymatic glycation.42

Thickening of the intima of epineurial arteries, due primarily to proliferation of endothelial cells, has also been observed in diabetic neuropathy43 but the degree of thickening was not correlated with the severity of neuropathy as assessed by the amount of nerve fiber loss.

Vascular Permeability

There have been conflicting reports of the permeability of endoneurial blood vessels in experimental diabetes. The present evidence suggests an increased permeability for small molecules46 but not for larger molecules. For human diabetic neuropathy an increased concentration of plasma macromolecules, including albumin and immunoglobulin, has been found in the endoneurium.49,50 On the other hand, Patel et al.51 have recently shown that permeation of glycated albumin into nerve is significantly greater than for native albumin and this may be at least part of the explanation, rather than any abnormality of the vessels themselves.

Animal Models

Tuck et al.,52 in streptozotocin-diabetic rats, found that endoneurial oxygenation and blood flow were reduced in the sciatic nerve and that vascular resistance was increased in the absence of any significant change in systemic blood pressure. The reduction in nerve conduction velocity that is found in this model can be partially reversed by rearing the animals in hyperbaric oxygen.53 The relevance of these findings to human diabetic neuropathy is speculative as significant degenerative changes with axonal degeneration and demyelination do not develop in this model, as demonstrated by Sharma and Thomas54 and others. Moreover, Zochodne and Ho,55 while confirming reduced intraneural oxygenation in streptozotocin-diabetic rats, failed to find evidence of diminished endoneurial blood flow.

Neuropathy Related to Chronic Respiratory Insufficiency

The occurrence of neuropathy in patients with respiratory insufficiency has been adduced as supportive evidence that hypoxia is responsible for diabetic neuropathy. Appenzeller et al.56 reported the presence of a distal polyneuropathy in 7 out of 8 patients with advanced chronic obstructive respiratory disease. The role of hypoxia in the explanation of neuropathy was difficult to assess in view of severe accompanying weight loss and malnutrition. Narayan and Ferranti57 and Faden et al.58 later confirmed the occurrence of neuropathy in patients with chronic respiratory insufficiency, and Vila et al.59 found electrophysiologic evidence of an axonopathy. Changes in neural blood vessels have also been described.60 It seems likely that a mild neuropathy may develop, for reasons not yet established, in patients with chronic respiratory insufficiency, yet its relevance to diabetic neuropathy is dubious. McLeod and Pollard61 concluded that it "seems to be of little clinical importance and so far has..."
Metabolic Hypotheses

Insulin Deficiency

Peripheral nerve, like the central nervous system, is not dependent on insulin for the regulation of glucose uptake or energy metabolism. This has been attributed to the presence of the blood-nerve and blood-brain barriers. Autonomic and sensory ganglia lack an efficient blood-nerve barrier and the question therefore arises as to whether they may be insulin-dependent. We have failed to detect any influence of insulin on glucose or amino acid uptake by dorsal root ganglia (N. Patel, D. Wright and P.K. Thomas, unpublished results). Any effects of insulin deficiency or tissue unresponsiveness would therefore have to be by less direct methods.

Sorbitol Accumulation

A major metabolic disturbance in peripheral nerve, in most of the experimental models and in man, is the accumulation of sorbitol and fructose in peripheral nerve. This is related to increased flux in the polyol pathway consequent upon saturation of the hexokinase shunt, with resultant accumulation of glucose in the nerve. In the polyol pathway, glucose is converted to sorbitol by aldose reductase and sorbitol to fructose by a polyol dehydrogenase. Evidence that this may be involved in the production of diabetic neuropathy is derived from observations on the use of aldose reductase inhibitors to prevent the accumulation of sorbitol in nerve. It is now conclusively documented that both from animal and human studies that nerve conduction velocity can be improved by aldose reductase inhibitors. Only modest evidence of improvement has been shown in clinical studies, but for the reasons given in the Introduction, substantial improvement is probably an impossible goal in treatment trials. Further long-term trials to establish whether deterioration can be prevented are merited. Encouraging evidence that aldose reductase inhibitors may increase regenerative sprouting of axons and a greater degree of remyelination has been provided by Sima et al. in serial sural nerve biopsies performed before and after 1 year of treatment with an aldose reductase inhibitor.

Myo-Inositol Depletion/Reduced Na⁺K⁺-ATPase Activity

A further metabolic abnormality identified in peripheral nerve in experimental diabetes in animals is a reduction of myo-inositol content. This has been linked by Greene and his coworkers to reduced Na⁺K⁺-ATPase activity, this observation having originally been made by Das, et al. In a series of studies in streptozotocin-diabetic rats, Greene and his collaborators found that myo-inositol deficiency in nerve to the occurrence of neuropathy, but this has largely been based on changes in nerve conduction velocity as an indicator of neuropathy. It was shown that both the deficit in conduction velocity and the reduced Na⁺K⁺-ATPase activity could be improved by dietary myo-inositol supplementation in the absence of any change in the severity of the hyperglycemia. There are difficulties, however, in accepting this hypothesis unreservedly. The relationship between myo-inositol concentrations in nerve and Na⁺K⁺-ATPase activity is not a simple one. In experimental galactose-induced neuropathy, Na⁺K⁺-ATPase activity is increased instead of being reduced, despite there being a reduced myo-inositol concentration in nerve. A major obstacle in applying the hypothesis to human diabetic neuropathy is the fact that myo-inositol concentrations are not reduced in human diabetic neuropathy. It is possible, however, that a small pool of phosphoinositides, derived from myo-inositol, is reduced, and that measurements of total myo-inositol concentration may not reflect this.

Altered Protein Synthesis and Axonal Transport

The uptake of amino acids and their incorporation into protein is reduced in dorsal root ganglia in streptozotocin-induced diabetic rats. Slow axonal transport has been shown to be impaired; both the velocity of slow component a (Sca) and the amount of material translocated in Sca have been found to be diminished. Structural proteins, including neurofilaments, are transported in Sca. They are largely responsible for determining axonal caliber. The reduction in the experimental animal is relatively small and the application of this finding to human diabetic neuropathy is uncertain. Although axonal atrophy has been found by some authors, it was not detected by Sugimura and Dyck or by Llewelyn et al.

Nonenzymatic Glycation

Hyperglycemia leads to the nonenzymatic glycation of proteins. No deleterious effects of this process on neuronal, axonal or Schwann cell function have so far been demonstrated, but it could contribute to vasculopathy.

Neurotrophic Effects

The clinical features of the common diabetic sensory and autonomic polyneuropathies are suggestive of a neuronal system degeneration that has a selective effect on particular sets of neurons. Pathologically, a distal 'dying-back' axonopathy has been demonstrated. Peripheral nerve has been shown to possess insulin receptors, the function of which is uncertain, as peripheral nerve, like the central nervous system, does not depend upon insulin for energy metabolism. It has been suggested that in the CNS insulin receptors are involved in longer term neurotrophic effects related to the maintenance of cell structure and integrity. It is conceivable that diabetic polyneuropathy could be on this basis, but a selective effect on peripheral nerve would have to be explained.

Immunologic Mechanisms

The possibility that immunologic mechanisms are involved in severe diabetic autonomic neuropathy has been raised by the finding of inflammatory infiltrates in autonomic ganglia of cases that have come to autopsy. The presence of antisym pathetic ganglion antibodies has also been demonstrated in patients with insulin-dependent diabetes, and is correlated with a diminished catecholamine response on standing.

Conclusions and Synthesis

A wide variety of mechanisms has been advanced as possibly being responsible for diabetic neuropathy. They should not be viewed as being mutually exclusive. The wide spectrum of clinical manifestations in the peripheral nervous system that may occur in patients with diabetes mellitus is a strong pointer...
towards the existence of multiple causal factors. It seems likely that in older patients in whom focal and multifocal neuropathies are more frequent, vascular factors will be more important. In the younger patients in whom a symmetric polyneuropathy is the usual presentation, there is current argument as to whether this is also vascular in origin, or whether it has a more direct metabolic basis. The writer favors the latter possibility, but the precise nature of the metabolic disturbance still eludes discovery.

REFERENCES