MINIREVIEW

Aging and Insulin Secretion (43879B)

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Abstract. **Aging in mammals has often been associated with decreased insulin secretion and a subsequent deterioration in the ability to maintain glucose homeostasis. However, recent studies have demonstrated that factors such as disease, obesity, and physical activity more closely reflect diminished insulin secretion rather than aging *per se*. Thus, the purpose of this article is to review recent studies of how biological aging, i.e. the process independent of disease states such as type II diabetes, may affect insulin secretion. To this end, this review will address the impact of aging on insulin secretion in terms of *in vivo* and *in vitro* assessment, as well as possible age-related alterations in the hormonal and neural regulation of insulin secretion. Finally, this review describes some evidence that alterations in the functional heterogeneity of the β-cell population may represent a means by which the endocrine pancreas is able to maintain appropriate insulin secretion during senescence.** [P.S.E.B.M. 1995, Vol 209]

Age-related alterations in various endocrine processes may have serious consequences with respect to the maintenance of homeostasis and can result in cellular senescence. The observation that serum glucose and insulin concentration increase with age has led some to suggest that a deterioration of glucose homeostasis is a normal consequence of aging and may even explain, in part, senescence. Because insulin plays a key role in the maintenance of glucose homeostasis, the effect of aging on insulin secretion has been the subject of extensive research.

The purpose of this article is to review recent studies of how biological aging (i.e., the process independent of disease states such as type II diabetes) may affect insulin secretion. To this end, this review will first briefly address the impact of aging on insulin secretion in terms of *in vivo* and *in vitro* assessment. Next, possible age-related alterations in hormonal and neural regulation of insulin secretion will be discussed. Finally, we will discuss a possible compensatory mechanism of the β-cell and/or islet of Langerhans that may allow the aging animal to maintain appropriate insulin secretion during senescence.

In Vivo Assessment of Insulin Secretion

The results of investigations using *in vivo* methods (e.g., oral and intravenous glucose administration) to assess the effect of aging on insulin secretion have been inconsistent. Several reports have documented significant age-related diminution of insulin secretion (1–3), whereas other studies have been unable to detect attenuation in the secretory response (4–9). For example, Kahn *et al.* (1) clearly demonstrated a higher and more sustained peak of serum glucose, indicative of attenuated insulin secretion, following an oral glucose tolerance test in older versus younger subjects. On the other hand, Bourey *et al.* (4) utilized a hyperglycemic clamp technique, in which blood glucose was maintained for 3 hr at 10 m*M*, to investigate the insulin secretory response of young (24 years) and older (65 years) subjects. Insulin secretion did not decrease significantly in older subjects with normal glucose tolerance.

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0037-9727/95/0000-0000$10.50/0

The inconsistent results among investigations using *in vivo* techniques to assess insulin secretion most likely reflects the inability of these methods to measure β-cell insulin secretion directly. This suggestion is supported by the observation that serum insulin concentration of older subjects does not, in all cases, correlate with *in vivo* measurement of prehepatic insulin secretion. To overcome the possible confounding factor of differential hepatic insulin metabolism between young and old, Pacini *et al.* (10) and Beccaro *et al.* (11) used C-peptide (a cleavage product of proinsulin that is secreted in equimolar amounts with insulin but is not metabolized by the liver) as a marker for prehepatic insulin release. These investigators independently reported similar serum glucose and insulin concentration of older versus younger humans, but the concentration of C-peptide in the elderly was much lower than in the young subjects, indicating impaired β-cell function and/or enhanced C-peptide clearance. The results of these two investigations imply that alterations in hepatic insulin metabolism may confound conclusions based on *in vivo* assessment of insulin secretion and mask possible age-related differences.

Additional difficulties may arise with interpretation of data from *in vivo* assessment of insulin secretion in aging subjects if obesity and level of physical activity are not considered (1–3). Several investigations have demonstrated that insulin secretion during *in vivo* "clamp" and "tolerance" tests does not decline in nonobese, healthy, or physically active older humans. Indeed, extensive research by Reaven and colleagues (12–15) has recently led these investigators to conclude that aging per se has a relatively minor effect on indices of glucose homeostasis, including insulin secretion. Rather, it is more likely that the age-related attenuation of insulin secretion observed in some investigations more closely reflects obesity and/or physical inactivity than does biological aging *per se*.

In Vitro Assessment of Insulin Secretion

Although recent investigations suggest that insulin secretion in humans does not decline as a function of biological aging, lack of control for numerous confounding variables during *in vivo* assessment of β-cell response has left many questions unanswered. To gain a better understanding of the effects of aging on insulin secretion and to control more precisely for confounding variables, many investigators have turned to *in vitro* assessment using whole perfused pancreas or islets of Langerhans isolated from rodents. However, similar to results from *in vivo* assessment of insulin secretion in humans, results from *in vitro* methods have been inconsistent. Some have found decreased (3, 16–22), increased (23, 24), or unchanged (25–29) insulin secretion with age. The various results among *in vitro* studies of insulin secretion most likely reflect

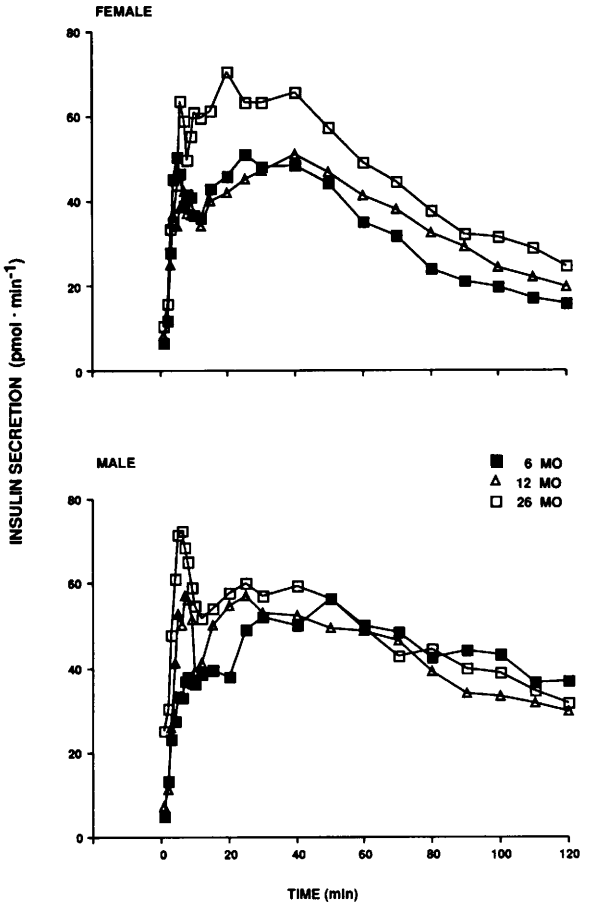

Figure 1. Mean glucose-stimulated (11.1 m*M*) insulin secretion of female and male Fischer 344 rats as determined using the whole perfused pancreas technique. For ease of reading, standard error bars are not included. (From Ruhe et al., 1992 [27]; copyright, The American Physiological Society.)

differences in age groups used as comparison for aging rats as well as differences in the definition of senescence in rodents.

The importance of defining senescence precisely in rodents when investigating age-related alterations is evident from the work of Reaven and colleagues (21, 30). These investigators suggested an age-related attenuation of insulin secretion per β-cell when they reported differences between 2- and 12-month-old Sprague-Dawley rats. Given that the median life span of the Sprague-Dawley rat is 26–28 months, 12 months of age would represent only young adulthood. Conversely, a 2-month-old Sprague-Dawley rat is growing at an exponential rate, indicating that this age group more closely models development rather than adulthood. When more developmentally mature rats and true senescent rats (defined as an age beyond the median life span for the particular strain) are compared, differences in insulin secretion have not been reported. For example, our laboratory has completed several investigations using whole perfused pancreas, and isolated islets and β-cells, from 6- (adolescent), 12- (young adult), and 26- (senescent) month-old Fischer

344 rats to describe the effects of aging on insulin secretion (25–27, 31). In all our investigations, we have not found significant differences in insulin secretion between the age groups (for example, see Fig. 1). Our investigations, as well as those by Starnes *et al.* (29), have led us to conclude that insulin secretion does not decline significantly with biological aging.

The conclusion that insulin secretion does not decline with biological aging does not, however, preclude the possibility that significant alterations in islet morphology, the extrinsic regulation of insulin secretion, or β-cell metabolism may occur with age. For example, Reaven *et al.* (21) observed that insulin secretion per β-cell, but not total insulin secretion, was significantly attenuated in older versus younger rats. Furthermore, pancreata isolated from older (18 months) Sprague-Dawley rats contained larger islets, more β-cells per islet, and more insulin per β-cell compared with younger rats (32). These findings are consistent with those of Adelman and colleagues (33), who found a greater proportion of larger islets in pancreata isolated from older versus younger rats. Larger islets secrete more insulin than do smaller islets regardless of the age of the rat. Based on these findings, Adelman (33) suggests that possible alterations of insulin secretion during aging may be overcome by the capacity of older animals to expand their pool of β-cells. This suggestion is consistent with our data (27) and implies that the insulin secretory function of the endocrine pancreas as a whole does not become impaired with age, but rather adapts to a changing metabolic environment.

Aging and Hormonal Regulation of Insulin Secretion

The hypothesis that insulin secretion is not impaired with aging but more closely reflects compensatory changes in response to the senescent metabolic environment implies possible alterations in the extrinsic, particularly hormonal, regulation of insulin secretion. Islets of Langerhans consist primarily of α, β, and δ cells which secrete glucagon, insulin, and somatostatin, respectively. Insulin secretion is stimulated by glucagon and inhibited by somatostatin (34–37). Because the cellular organization of the islet places the α and δ cells in close proximity to the β cell, it is reasonable to predict that the secretion of glucagon and/or somatostatin will have a significant effect on insulin secretion, referred to as paracrine regulation. It is possible, then, that an age-related change in the relative proportion of α and/or δ cells, or loss of secretory capacity of these cells, could alter insulin secretion. The suggestion that age-related changes in paracrine regulation lead to altered insulin secretion is consistent with the observations of decreased density, mean area, and secretion of aging glucagon-containing α cells (38, 39). Casad *et al.* (40), using the whole perfused pancreas technique, observed that the inhibitory action of somatostatin on insulin secretion is increased with age (40). However, the finding that insulin secretion during aging is altered through paracrine regulation has not been supported by others. For example, Magal *et al.* (36) and Starnes *et al.* (29) did not report alterations in insulin secretion with age or differences in the regulatory influence of somatostatin on insulin secretion among younger and older rats. Adelman *et al.* (33) have suggested that the differences among some of these findings may reflect the size of islets used to investigate insulin secretion. Smaller islets of older versus younger animals are apparently more sensitive to the action somatostatin and thus secrete less insulin than do larger islets. Investigations reporting greater inhibitory action of somatostatin in aging rats may have used a disproportionate number of smaller islets. Regardless, Chaudhuri *et al.* (18) and Adelman (33) have suggested that the greater proportion of larger islets reported in the pancreata of aged rats is an adaptation that allows these animals to maintain insulin secretion.

Although glucagon and somatostatin represent the majority of noninsulin hormones secreted by the islet, recent evidence suggests that other peptides synthesized within islets can affect insulin secretion. For example, islet amyloid polypeptide has been shown to be localized in the β-cell and is believed to be cosecreted with insulin and regulated by the same mechanisms that affect insulin secretion (41). Neuropeptide-Y, also synthesized within islets, has been suggested to be an important intra-islet paracrine hormone (42). Consequently, the effects of islet amyloid polypeptide and neuropeptide-Y on insulin secretion have been investigated separately in various species, with conflicting and sometimes ambiguous results. In some studies, decreases in glucose-stimulated insulin secretion have been reported (42–44), while others have suggested that these peptides have little effect on insulin secretion (45–48). Although it could be hypothesized that an age-related increase in islet amyloid polypeptide and neuropeptide-Y release may function to alter insulin secretion, this possibility has yet to be investigated.

Investigations concerned with the effects of aging on insulin secretion have also considered how age-related changes in other secretory tissues extrinsic to the pancreas may influence insulin secretion. Diminished gastrointestinal function, previously reported to occur with age (49–51), may alter the secretion of gut hormones, such as gastric inhibitory peptide, enteroglucagon, gastrin, secretin, and cholecystokinin, which in turn may effect insulin secretion. Groop (52) used gastric inhibitory peptide as a model to examine the relationship between insulin secretion, gastrointestinal hormones, and aging. This investigator reported

that gastric inhibitory peptide did not differ between younger and older individuals yet insulin secretion was significantly greater in the elderly subjects. These findings suggest that the secretion of gastrointestinal hormones do not decline with age nor do they have a significant impact on insulin secretion. While investigations into the age-related effects of other gastrointestinal hormones have been limited, the data of Groop and others (53) suggest that the contribution of gut hormones to possible alterations on insulin secretion in the aged may be small.

Aging and Neural Regulation of Insulin Secretion

The islets of Langerhans are richly innervated with both parasympathetic and sympathetic divisions of the central nervous system. Stimulation of the sympathetic nervous system inhibits insulin secretion, while parasympathetic nervous system stimulation potentiates insulin secretion. Although the importance of neural regulation of insulin secretion remains controversial, its relationship to aging may be significant since several investigations have demonstrated an age-associated increase in sympathetic nervous system activity (54–56). Young *et al.* (57) who reported that serum norepinephrine, the major neurotransmitter of the sympathetic nervous system, is greater in older humans during an oral glucose tolerance test compared to younger subjects. Curry and McDonald (58) suggest that the enhanced sympathetic tone observed in aging humans during glucose loads may reflect a compensatory mechanism that works to prevent hyperinsulinemia. Using an *in situ* innervated perfused pancreas preparation in young (5 months) and old (26 months) rats, these investigators reported that insulin secretion did not differ between age groups when the neural tracts were intact. Ablation of the neural tract significantly increased insulin secretion of the older animals compared to the younger rats and nonablated younger and older rats, suggesting inhibitory control of insulin secretion in the older rats. On the other hand, stimulatory control of insulin secretion was demonstrated in younger rats as secretion declined significantly following ablation of the neural tract compared with older animals. Together, these results indicate that as the animal ages, neural regulation of insulin secretion "shifts" from stimulatory in younger rats to inhibitory during senescence. The "compensatory" mechanism that prevents hyperinsulinemia in the older animal does not reflect greater sensitivity to norepinephrine (31). These results are consistent with the suggestion that neural regulation of insulin secretion during aging is altered through greater sympathetic tone.

Although the mechanism associated with the enhanced sympathetic nervous system-mediated inhibition of insulin secretion in the older animal has not yet been fully elucidated, some have suggested that the inhibition reflects a complex interaction between the α- and β-adrenergic receptors of the β-cells. The inhibitory actions of catecholamines on insulin secretion are mediated principally through the α-adrenergic receptor (59). However, some evidence exists suggesting that epinephrine also enhances insulin secretion during stress and/or hyperglycemia via β-adrenergic stimulation (60). It is possible, therefore, that an age-related decrease in sensitivity to epinephrine, in terms of the β-cell's β-adrenergic receptor, would result in relatively unopposed α-adrenergic stimulation during stress state (i.e., a relative decrease in insulin secretion). This is a reasonable hypothesis since numerous studies have demonstrated diminished β-adrenergic receptor mediated function in several tissues in both aged humans and animals (61–63). However, Morrow *et al.* (64) have reported that insulin secretory responses to β-adrenergic receptor stimulation in young (20–25 years) and old (62–74 years) human subjects did not differ significantly. These findings indicate that insulin secretory response of the β-cell to β-adrenergic stimulation is not diminished in the elderly, suggesting that sensitivity to epinephrine is not altered with age (64). In a subsequent investigation, Morrow and colleagues (65) assessed the responses of insulin secretion and insulin action to increasing physiological doses of epinephrine in both young (19–26 years) and older (62–75 years) adults. The ability of epinephrine to inhibit glucose-induced insulin secretion, and the dose-response relationship between epinephrine and insulin action, were similar in young and old subjects. Together, these data suggest that β-cell sensitivity to catecholamines does not decline with age.

Critical gaps remain in our understanding of how hormonal and neural regulation effect insulin secretion, particularly with respect to aging. The investigations completed to date, although not conclusive, strongly imply that the action of these two regulatory systems does not result in diminished insulin secretion during aging. Rather, data are beginning to accumulate suggesting that hormonal and neural regulation of β-cell function during aging act to compensate for alterations in the metabolism of the senescent animal. In turn, this "compensatory regulation" helps to maintain appropriate insulin secretion during aging.

Aging and Insulin Biosynthesis

There is little evidence to suggest that any age-related changes in β-cell function occur at the level of transcription. Microscopic analysis of 30-month-old rat β-cell nuclei reveals that with age, the relative volume of the condensed (nontranscribable) chromatin increases progressively at the expense of the dispersed (transcribable) form, suggesting that transcriptional activity could be reduced in these aged cells (66). It was noted in this study, however, that under nonphys-

iological *in vitro* conditions, old β-cell nuclei are able to react in the same way as young ones with respect to chromatin redistribution and nuclear size increase, suggesting that the machinery for transcription remains fully functional in the older cells. In a study of isolated islets from young and old Fischer rats (67), it was determined that the levels of preproinsulin mRNA, the direct product of transcription, did not change with age. These data strongly suggest that any alteration in the insulin synthesis/secretion pathway is post-transcriptional.

In studies using the isolated islet preparation (67), it was demonstrated that although preproinsulin mRNA levels remained the same, glucose-stimulated proinsulin biosynthesis was decreased in islets of old animals, suggesting impaired translation. It was not determined if the impairment was due to a defect in the signaling mechanism or to degeneration of the biosynthetic apparatus. Evidence for the latter was provided by DeClercq *et al.* (66) who observed, in freshly isolated islets, a decrease with age in the volume density of rough endoplamic reticulum and Golgi complex, changes that could affect proinsulin biosynthesis. However, total insulin content of islets has been shown not to decline with age, suggesting no alteration in biosynthetic capacity (27, 67, 68). Altogether, current data do not provide enough evidence to draw any sound conclusion regarding the existence of age-related changes in β-cell function at the level of biosynthesis.

Aging and the Stimulus-Secretion Coupling Mechanism of Insulin Secretion

The preponderance of evidence now suggests that postdevelopmental aging *per se* does not result in significantly decreased insulin secretory capacity of the whole endocrine pancreas. As discussed above, extrinsic regulation of insulin secretion (i.e., hormonal and neural regulation) appears to be compensatory. However, it is also possible that changes intrinsic to the β-cell are equally responsible for maintaining insulin secretion in the aging animal.

Within the β-cell, insulin secretion is dependent upon the stimulus-secretion coupling mechanism, the series of events that link the rise in serum glucose concentration to insulin secretion. This mechanism is unique among the endocrine tissues in that the principal agonist, glucose, produces its effect by its own oxidation (Fig. 2). Glucose oxidation in the β-cell generates ATP and increases the intracellular ATP/ADP ratio, which is most likely responsible for the closure of the ATP-sensitive potassium channels (69–71) and subsequent membrane depolarization (72). Voltage-dependent calcium channels open in response to membrane depolarization, allowing an influx of extracellular calcium necessary for insulin secretion to occur.

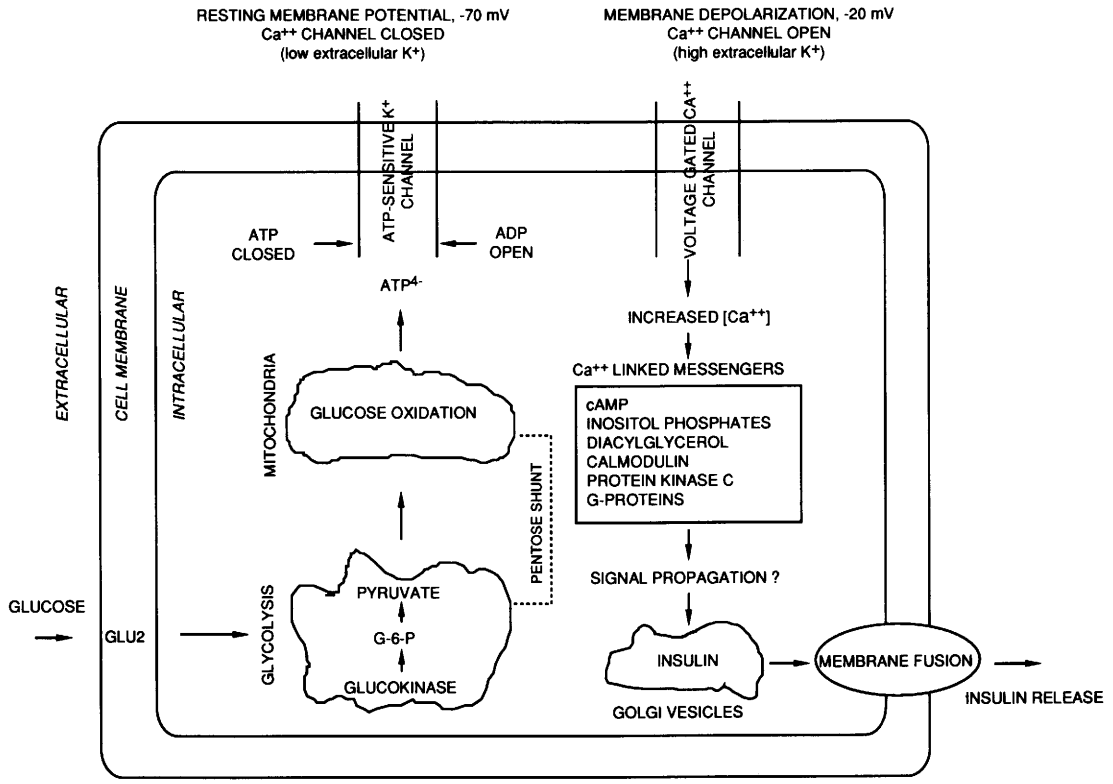

Figure 2. Schematic representation of the stimulus-secretion coupling mechanism of the β-cell. Arrows indicate direction of the signal. (From Ruhe RC, McDonald RB. Aging, insulin secretion, and cellular senescence. In: Watson RR, Ed. Handbook of Nutrition in the Aged. (2nd ed). Boca Raton: CRC Press, pp 73–98, 1992; copyright, CRC Press, Inc.)

Because glucose oxidation and ion channel activity are well established as crucial to insulin secretion, most investigations of the affects of aging on the stimulus-secretion coupling mechanism have focused on these events.

Glucose Oxidation, Insulin Secretion, and Aging

The close relationship between glucose oxidation and insulin release (73, 74) has influenced several investigators to examine the affects of aging on β-cell glucose oxidation. Investigations by Elahi *et al.* (75), Reaven and Reaven (76), Sartin *et al.* (77), and Castro (17), have demonstrated attenuated glucose oxidation concomitant with diminished insulin secretion in islets isolated from older versus younger rats. In contrast, Ammon *et al.* (78) reported no differences in glucose utilization nor in NADPH production in islets from old rats as compared to islets from younger animals. Burch *et al.* (23) observed an increased glucose oxidation rate which was associated with enhanced glucokinase, phosphofructokinase, and glucose-6-phosphate dehydrogenase activity in islets of 18-month-old rats compared with 8-month-old animals. A similar increase in glucose oxidation was found by Leiter *et al.* (24) of islets isolated from the C57BL/6J mouse. In a recent study using islets of Langerhans isolated from male Fischer 344 rats, Ruhe *et al.* (27) observed slightly greater rates of glucose oxidation in islets of older (26 months) animals as compared to islets from younger (6 months) animals. Having demonstrated that glucose sensitivity of the islets is not decreased with age, these researchers proposed that any age-related alteration in the stimulus-secretion coupling mechanism that may serve to maintain glucose-stimulated insulin secretion is at a step distal to glucose oxidation.

β-Cell Ion Channel Activity and Aging

In the β-cell, as in other excitable cells, the resting membrane potential is determined by the transmembrane K^+ gradient and the degree of K^+ permeability, which in the β-cell is regulated by the presence of ATP-sensitive K^+ (K_{ATP}) channels. There remains some debate about the precise coupling between substrate metabolism and K_{ATP} channel closure, although it is currently thought that this is predominantly mediated through alterations in the cytoplasmic ATP/ADP ratio (79, 80). In any event, it is evident that ion movements and changes in membrane potential are critical to the insulin secretory process, so alterations in ion channel activity with age would almost certainly affect insulin secretion. Studies of ion movements in human and mice lymphocytes (81, 82), as well as neurons of various animals models (83–85), have demonstrated an age-related decrease in K^+ channel responsiveness and/or decreased membrane potential. Some authors suggest that changes in membrane phospholipid composition and lipid fluidity may modify the opening of these channels, modifications which may be associated with the noted age-related dysfunction of the immune system and altered neuronal functions. It is thus reasonable to examine the possibility that similar modifications may occur in all excitable cells, including the β-cell. Using male Wistar rats, Ammon *et al.* (86) observed attenuated K^+ movement, as measured by $^{86}Rb^+$ efflux, and decreased insulin secretion in response to glucose stimulation of islets isolated from 24- vs. 3-month-old animals. The authors concluded that the capacity of the β-cell to inhibit K^+ efflux in response to glucose is decreased in old age. The implications of this study are unclear, however, as a developmentally mature age group was not included.

Islet glucose oxidation is related directly to ATP production, and intracellular ATP/ADP ratio is related directly to K^+ channel closure. Consequently, if glucose oxidation is maintained in islets of aging animals, as implied from the studies discussed previously, then any change in the responsiveness of K_{ATP} channels is likely due to ATP sensitivity of the K_{ATP} channels, rather than to a decreased availability of ATP. Draznin *et al.* (87) investigated this possibility using glyburide, a substance that closes K_{ATP} channels in a manner similar to ATP but independent of glucose oxidation, and observed similar rates of insulin secretion between islets of older and younger F344 rats. Consistent with these data, Ruhe *et al.* (28) used islets isolated from 6-, 12-, and 26-month-old male F344 rats and found that insulin secretion of islets incubated in the presence of a nonstimulatory (1.7 m*M*) or a stimulatory (11.1 m*M*) concentration of glucose and glyburide did not differ significantly among age groups. Taken together, these studies imply that if an adaptive alteration in the β-cell stimulus-secretion coupling mechanism occurs in the older rat, it does so at a step distal to glucose oxidation and K_{ATP} channel-mediated response.

Calcium and Second Messengers of Insulin Secretion

The mechanisms of stimulus-secretion coupling requiring increased intracellular calcium are not completely understood, and the effects of age on these mechanisms are even less clear. It is generally accepted that an increase in intracellular free calcium concentration ($[Ca^{2+}]_i$) is a crucial event in the triggering of insulin release by nutrients (88–91). Furthermore, the increase in $[Ca^{2+}]_i$ occurs via the influx of Ca^{2+} through voltage-gated calcium channels as demonstrated with rodent islets (92, 93), human islets (94), and in single isolated β-cells (90, 95–97).

There is evidence to suggest that increased $[Ca^{2+}]_i$ from the influx of extracellular calcium activates calcium-calmodulin-sensitive enzymes (98–100) and pro-

tein kinase (PKC) (101–103), which have been reported to promote insulin release from the β-cell. Moreover, intracellular free calcium, in concert with other second messengers (diacylglycerol, cAMP and/or as yet uncharacterized modulators), may initiate the phosphorylation of key proteins involved in exocytosis. However, several recent studies have indicated that pKC (104–106), diacylglycerol (107), and calmodulin (108) may not be important mediators of glucose stimulated insulin secretion. The work of Yaney *et al.* (108) appears to support the suggestion of previous investigators that cAMP/calmodulin play a role in amplifying stimulus-secretion coupling and are not essential to basal function. Similarly, Ashcroft *et al.* (109) have proposed that protein kinase activation potentiates insulin secretion indirectly by sensitizing the secretory mechanism to intracellular calcium and by promoting amplification of the secretory response. Additionally, Arkhammar *et al.* (110) recently demonstrated that PKC may be tonically active and effective in the maintenance of the phosphorylation state of the voltage-gated calcium channels, enabling an appropriate function of these channels in the insulin secretory process.

Given the importance of $[Ca^{2+}]_i$ to insulin secretion, it has been hypothesized that the maintenance of insulin secretion by the aging endocrine pancreas reflects an alteration in β-cell regulation of $[Ca^{2+}]_i$. Using single-cell microspectrofluorimetry to examine differences in $[Ca^{2+}]_i$ among individual β-cells, our laboratory has recently observed that the change in $[Ca^{2+}]_i$ upon glucose stimulation did not differ significantly between β-cells from young and old rats (unpublished results). These data suggest that the maintenance of insulin secretion by the senescent animal cannot be attributed to alterations in the regulation of $[Ca^{2+}]_i$ by the β-cell.

Aging and β-Cell Response Heterogeneity

It is now well recognized that the secretory response of the pancreatic β-cells to glucose stimulation is heterogeneous in nature (111–113). That is, in any given population of isolated β-cells, only about 70% of the cells are responsive to glucose in terms of insulin secretion. Direct evidence of functional heterogeneity among β-cells comes from autoradiographs of isolated rat islet β-cells in which newly synthesized proteins had been labeled at different glucose concentrations (114). In addition, numerous experiments with single β-cells indicate the existence of intercellular differences in responsiveness to glucose (111, 113, 115–118). Given these recent findings, we hypothesized that the ability of the senescent animal to maintain insulin secretion reflects altered functional heterogeneity of the pancreatic β-cell population. That is, in order to maintain insulin secretion during aging, more β-cells in older versus younger rats are responsive to glucose. Our laboratory has now completed preliminary investigations that support this hypothesis. We compared the responsiveness to glucose of β-cells isolated from 6-, 12-, and 26-month-old rats, with responsiveness defined as a change in $[Ca^{2+}]_i$ of at least 50% greater than $[Ca^{2+}]_i$ at the nonstimulatory glucose concentration of 5.5 m*M*. Approximately 76% of the β-cells isolated from the 26-month-old rats were responsive to glucose at concentrations ≥7.5 mM. For the 6- and 12-month-old animals, approximately 63% and 65%, respectively, of their β-cells were responsive to the stimulatory glucose concentrations (Table I). These results suggest that in the senescent rat, the secretory capacity of the islet is maintained, in part, through a change in the functional heterogeneity of the β-cell population (i.e., an increase in the percentage of β-cells that are responsive to glucose).

Summary and Conclusion

Aging in mammals has often been associated with decreased insulin secretion and a subsequent deterioration in the ability to maintain glucose homeostasis. However, recent studies using *in vivo* and *in vitro* techniques to assess glucose-stimulated insulin secretion have demonstrated that factors such as disease, obesity, and physical activity more closely reflect diminished insulin secretion rather than aging *per se*. In the absence of specific pathologies or environmental factors, biological aging is not associated with diminished insulin secretion.

The ability of the senescent animal to maintain insulin secretory capacity at a level comparable to that of younger animals is apparently the result of a com-

Table I. Responsiveness and Sensitivity to Glucose Stimuli of Pancreatic β-Cells Isolated from F344 Rats of Different Ages

Age (months)	Nonresponsive cells (% of all cells)	Responsive cells (% of glucose-responsive cells)	
		7.5 m*M* glucose	11.1 m*M* glucose
6 (n = 84)	36.9[a]	57.5[a]	46.6[a]
12 (n = 82)	35.4[a]	60.0[a]	40.0[a]
26 (n = 125)	24.0[b]	72.4[b]	27.6[b]

Note. Individual β-cells were grouped into three categories based on changes in $[Ca^{2+}]_i$ following glucose stimulation: Glucose-nonresponsive, responsive at 7.5 m*M* glucose, and responsive 11.1 m*M* glucose. The criterion for responsiveness was a >50% increase in $[Ca^{2+}]_i$ over basal $[Ca^{2+}]_i$ at 5.5 m*M* glucose. Within a column, values not sharing a common letter superscript are significantly different by chi-square test ($P < 0.05$) (Unpublished results).

plex interaction of factors extrinsic and intrinsic to the islets of Langerhans. Several studies have demonstrated that extrinsic regulators of insulin secretion (e.g., hormonal and neural) undergo age-related alterations that may compensate for physiologic and metabolic changes in the senescent animal. Age-related changes intrinsic to the islets may also play a role in maintaining insulin secretion. Although no functional changes specific to the β-cell have been observed, aging does appear to alter the functional heterogeneity among β-cells within the islet. That is, the percentage of β-cells that are glucose-responsive, as well as the glucose sensitivity of the responsive cells, were recently shown to be greater in islets of the senescent rat compared with islets of younger animals. It is possible that altering functional heterogeneity of the β-cell population may represent a means by which the endocrine pancreas is able to maintain appropriate insulin secretion during senescence.

Some of the work presented in this article was supported, in part, by National Institutes of Health Grants AG00429 and AG06665, and through a gift from The Sugar Association, Inc.

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