Neuroscience Vol. 37, No. 2, pp. 367–375, 1990
Printed in Great Britain

0306-4522/90 $3.00 + 0.00
Pergamon Press plc

SHORT-TERM GLUCOCORTICOID MANIPULATIONS AFFECT NEURONAL MORPHOLOGY AND SURVIVAL IN THE ADULT DENTATE GYRUS

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Abstract—In order to determine whether short-term glucocorticoid manipulations influence the morphology and survival of neurons in the adult mammalian hippocampal formation, we performed quantitative analyses of Golgi-impregnated and Nissl-stained tissue from the brains of sham operated male rats, adrenalectomized male rats and adrenalectomized male rats which received corticosterone replacement. Three days after adrenalectomy, massive cell death, as detected by a dramatic increase in number of pyknotic cells, was observed in the granule cell layer of the dentate gyrus. By seven days following adrenalectomy, the numbers of pyknotic cells were even greater. Moreover, significant decreases in cross-sectional cell body area and numbers of dendritic branch points of Golgi-impregnated dentate gyrus granule cells were detected at seven days after adrenalectomy. Replacement of corticosterone to adrenalectomized rats prevented the appearance of large numbers of pyknotic cells as well as the decrease in granule cell cross-sectional cell body area and the numbers of dendritic branch points. In contrast, no obvious signs of degeneration were detected in the pyramidal cell layers of the CA1 and CA3 regions of the hippocampus at either three or seven days following adrenalectomy. In addition, no significant changes in morphological characteristics were observed in CA1 or CA3 pyramidal cells with adrenalectomy.

These results show that dentate gyrus granule cells require glucocorticoids for their survival and for the maintenance of normal morphology and suggest that granule cell morphology and/or survival may undergo constant fluctuation in response to diurnal rhythms or stress-induced changes in glucocorticoid levels.

It is now generally accepted that the hippocampal formation is a major target for glucocorticoid action in the brain. This contention is supported by the presence of a high number of both Type 1 and Type 2 glucocorticoid receptors in this neural region in adulthood.[18,21] The effects of glucocorticoids on the hippocampal formation are of particular interest due to the proposed role of this structure in the processes of learning and memory[19,28] and neuroendocrine function.[24,33] Imbalances in the level of circulating glucocorticoids during both development and adulthood have been shown to alter hippocampal physiology[9,27,30,32] as well as processes associated with the hippocampus, such as learning and memory.[8,17,31] The cellular mechanisms underlying these physiological changes are not completely understood.

A number of studies which have aimed at understanding the cellular effects of glucocorticoid manipulations have employed long-term treatments, usually of the order of three to four months. For example, the detrimental effects of excess glucocorticoids administered chronically have been well documented. The results of these studies support the notion that high levels of glucocorticoids, administered experimentally or induced by stress, are toxic to hippocampal pyramidal cells.[23,29] On the other hand, recent evidence suggests that removal of circulating glucocorticoids by adrenalectomy results, over a three- to four-month period, in massive cell death of granule cells in the dentate gyrus.[27] These results suggest that glucocorticoids exert a trophic influence over this neuronal population. Such long-term treatments, however, make it difficult to determine whether glucocorticoids play a direct role in eliciting any observed changes. To date, no study has examined the effects of short-term glucocorticoid manipulations on morphology and survival of neurons in the adult dentate gyrus. In this study, we examined the effects of short-term (three- and seven-day) glucocorticoid manipulations on the morphologic characteristics and the survival of three different neuronal populations in the hippocampal formation: granule cells from the dentate gyrus, pyramidal cells from the CA3 region and pyramidal cells from the CA1 region.

EXPERIMENTAL PROCEDURES

Surgical procedures and treatments

Young adult male Sprague–Dawley (Charles River) rats (220–240 g, 58 days old) were used for this study. These rats were assigned to one of the following treatment groups: (1) sham operation under Metofane anesthesia, (2) adrenalectomy under Metofane anesthesia, or (3) adrenalectomy under Metofane anesthesia with corticosterone replacement (25 μg/ml) in the drinking water starting the day after surgery. This dose results in blood levels of corticosterone which are within the lower range of normal levels (unpublished observation). All adrenalectomized rats received 0.9% saline in the drinking water during the interval

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between adrenalectomy and perfusion. The experiment was performed with a seven-day interval between surgery and perfusion. In addition, sham operated and adrenalectomized rats were examined with a three-day interval between surgery and perfusion.

On the day of perfusion, rats were deeply anesthetized with Ketamine and transcardially perfused with 100–150 ml 4% paraformaldehyde in 0.1 M phosphate buffer with 1.5% (v/v) saturated picric acid. Brains were postfixed for 24 h in a solution of the same composition as the perfusate.

Histological procedures

The brains from the seven-day experiment were processed using a modified version of the single-section Golgi impregnation method.[11] Briefly, coronal sections 100 μm thick were cut by use of a Vibratome in a bath of 3.0% potassium dichromate in distilled water and stored in this solution for 24 h. Following this, the sections were rinsed in distilled water and mounted onto ungelatinized slides. Coverslips were glued over the tissue sections at the four corners and the slide assemblies were placed in 1.5% silver nitrate in distilled water for 24 h in the dark. The slide assemblies were then dismantled, the sections were rinsed in distilled water, dehydrated, cleared and coverslipped.

For both the three- and seven-day experiments, coronal sections 100 μm thick from the same brains were mounted onto gelatinized slides, stained for Nissl using a Cresyl Violet stain, dehydrated, cleared and coverslipped.

Data analysis

The slides containing Golgi-impregnated brain sections were coded prior to quantitative analysis. The code was not broken until after the analysis was completed. Camera lucida tracings (500 ×) were obtained from selected granule cells of the dentate gyrus and pyramidal cells of the CA3c region. In order to be selected for morphological analysis, Golgi-impregnated hippocampal cells had to possess the following characteristics: (1) location within the septal portion of the dentate gyrus for granule cells or the CA3c region for pyramidal cells; (2) relative isolation from neighboring impregnated cells in order to allow identification of dendrites which emanated from specific cells; and (3) dark and consistent impregnation throughout the extent of the neuron. In order to determine whether or not glucocorticoids affect one subpopulation of granule cells more than others, camera lucida tracings were performed for three types of dentate gyrus granule cells: (1) granule cells located in the genu of the dentate gyrus; (2) granule cells with a single primary dendrite (which are located primarily in the deep aspect of the granule cell layer[13]); and (3) granule cells with multiple primary dendrites (which are located primarily in the superficial aspect of the granule cell layer[13]). These two latter types of granule cells were always selected from the suprapyramidal blade of the dentate gyrus. For each brain, 24 neurons (six of each type) were analysed. A total of 360 Golgi-impregnated neurons were traced for this study.

In order to determine whether specific regions of the dendritic tree were affected by the experimental conditions, a concentric ring analysis, examining the amount of dendritic material at 10-μm intervals from the cell body, was performed for each camera lucida tracing.[26] The total number of dendritic branch points was also determined from the camera lucida tracings. In addition, the total length of the dendrites in a given dendritic tree detectable in a 100-μm section was determined from the camera lucida tracings by use of an image analysis morphometry program (Southern Micro Instrument Inc., Atlanta, GA).

The mean cross-sectional cell body area of granule cells in the suprapyramidal as well as infrapyramidal blades of the dentate gyrus and pyramidal cells of the CA3 and CA1 regions was also determined (1250 ×) by use of the Southern Micro Instrument morphometry program. A minimum of 50 cell body area measurements were obtained for each cell type for each brain. In addition, the density of specialized spines of CA3 pyramidal cell apical dendrites, called excrescences, were analysed. For each selected CA3 pyramidal cell, the apical dendritic shaft was drawn (1250 ×) using a camera lucida drawing tube, and the number of excrescences was determined for that length. The length of the apical shaft was determined by use of the Southern Micro Instrument morphometry program and the number of excrescences was expressed per 10-μm length. A total of 144 apical dendritic shafts (12 from each brain) were examined for this study. The means of these variables were determined for each animal and the data were subjected to a one-way ANOVA with Tukey HSD *post hoc* comparisons.

The slides containing Cresyl Violet stained brain sections were coded prior to analysis. The code was not broken until after the analysis was completed. Nissl-stained sections were examined for the number of pyknotic cells within the granule cell layer of the dentate gyrus and the pyramidal cell layers of the CA1 and CA3 regions. Pyknotic cells were characterized by their condensed chromatin, lack of a nuclear membrane and light or absent cytoplasm.[5,7,12] Cell counts of these degenerating profiles were made and templates of their distribution were mapped by use of a camera lucida drawing tube. The cell layers from each section were traced (50 ×) with the camera lucida drawing tube and the area of each cell layer was determined using the Southern Micro Instrument morphometry program. Means of the number of pyknotic cells per 10^6 μm^2 region were determined for each animal for the granule cell layer of the dentate gyrus and the pyramidal cell layers of the CA3 and CA1 regions. These values were analysed using a one-way ANOVA with Tukey HSD *post hoc* comparisons.

RESULTS

Effects of glucocorticoid manipulations on hippocampal morphology

Light microscopic examination of Golgi-impregnated tissue revealed reliable and consistent staining throughout the hippocampal formation for all treatment groups. Quantitative analysis of Golgi-impregnated dentate gyrus granule cells revealed significant overall differences in cross-sectional cell body area in both the suprapyramidal blade ($P < 0.005$) and the infrapyramidal blade ($P < 0.005$) with seven-day treatment (see Table 1). Seven-day adrenalectomy resulted in a significant decrease in cell body area of granule cells of both the suprapyramidal and infrapyramidal blades compared to sham operated animals ($P < 0.05$, see Fig. 1, Table 1). Replacement of corticosterone in the drinking water to adrenalectomized animals resulted in a prevention of the decrease in cell body area of granule cells from both blades of the granule cell layer (Table 1).

In addition to these observed changes in cell body area, granule cells of the three morphologic types examined (granule cells located in the genu, single primary dendrite granule cells and multiple primary dendrite granule cells) showed significant overall changes in the number of dendritic branch points with treatment [$P < 0.05$ for granule cells located in the genu (Fig. 2), $P < 0.05$ for single dendrite granule cells (Figs 1 and 2), $P < 0.025$ for multiple dendrite granule cells, see Table 1, Fig. 2]. Adrenalectomy

Table 1

Morphological variable	Sham operation $n = 5$	Adrenalectomy $n = 6$	Adrenalectomy plus corticosterone $n = 4$
Cell body area of granule cells located in the suprapyramidal blade (μm^2)	181.8 ± 2.7	145.7 ± 4.8*	171.9 ± 6.2
Cell body area of granule cells located in the infrapyramidal blade (μm^2)	196.9 ± 12.8	142.2 ± 4.4*	189.4 ± 7.3
Number of dendritic branch points of granule cells located in the genu	4.9 ± 0.9	2.9 ± 0.4*	4.2 ± 0.2
Number of dendritic branch points of single primary dendrite granule cells	6.4 ± 0.6	4.7 ± 0.3*	5.7 ± 0.4
Number of dendritic branch points of multiple primary dendrite granule cells	5.8 ± 0.5	4.2 ± 0.4*	6.2 ± 0.5
Length of dendrites within 100-μm section of granule cells located in the genu	493.8 ± 61.6	323.1 ± 33.2	392.1 ± 46.7
Length of dendrites within 100-μm section of single primary dendrite granule cells	653.1 ± 64.4	675.7 ± 42.8	759.5 ± 28.2
Length of dendrites within 100-μm section of multiple primary dendrite granule cells	926.0 ± 29.0	765.7 ± 76.4	839.6 ± 48.1
Number of pyknotic cells in dentate gyrus/10^6 μm^2	6.4 ± 1.0	2163.8 ± 556.0*	6.9 ± 2.2

Values represent mean ± standard error. These data were analysed by one-way ANOVA with *post hoc* comparisons (see text). Asterisks equal significant difference ($P < 0.05$) from sham operated.

resulted in a significant decrease in number of dendritic branch points ($P < 0.05$) in all three types of granule cells compared to the sham group. Replacement of corticosterone to adrenalectomized animals resulted in dendritic branch point values which were not significantly different from the sham group (Fig. 2). Concentric ring analysis did not reveal any significant differences in the amount of dendritic material in any portion of the dendritic tree ($P > 0.1$ for all comparisons). Rather, a trend toward a decrease in amount of dendritic material was observed throughout the extent of the dendritic tree. No significant differences in the overall length of the dendrites in a 100-μm section were observed for granule cells with single primary dendrites ($P > 0.25$), granule cells with multiple primary dendrites ($P < 0.25$) or granule cells located in the genu ($P < 0.25$; see Table 1).

Quantitative analysis of Golgi-impregnated CA3 pyramidal cells revealed no significant differences in the number of branch points in apical ($P < 0.25$) or basal ($P > 0.25$) dendritic trees, or in the overall

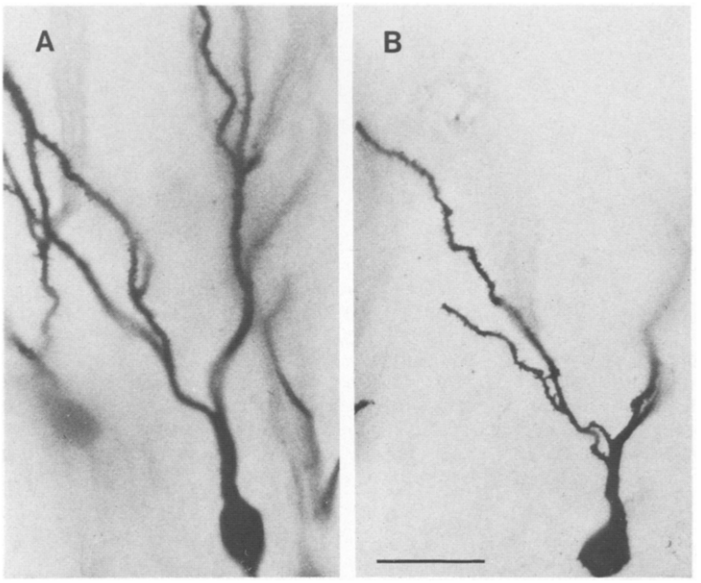

Fig. 1. Golgi-impregnated dentate gyrus granule cells with a single primary dendrite from brains of sham operated (A) and adrenalectomized (B) animals. Note the decrease in cell body area and dendritic branch points in B compared with A. Scale bar = 20 μm and applies to both frames.

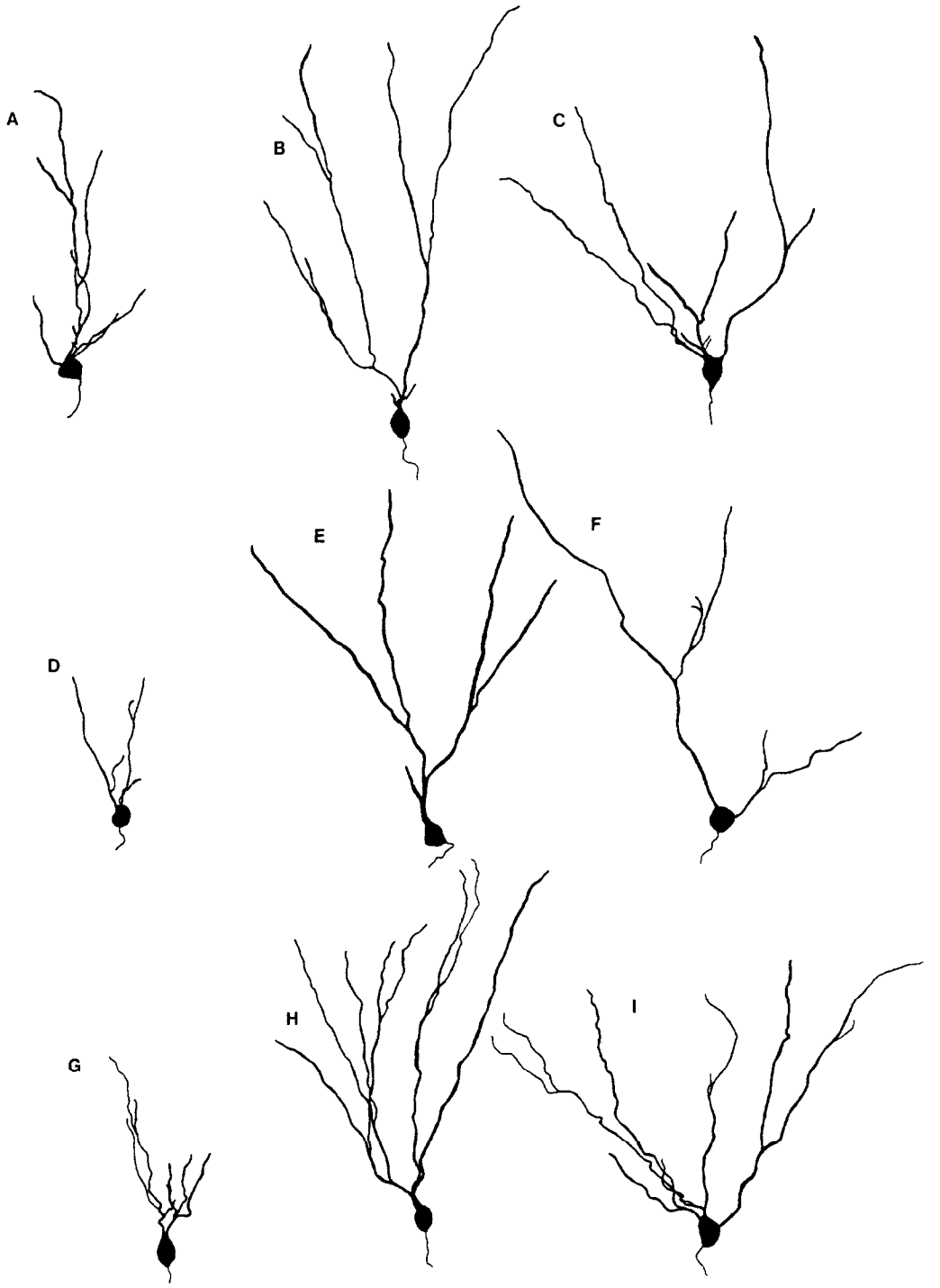

Fig. 2. Camera lucida drawings of representative Golgi-impregnated dentate gyrus granule cells from brains of sham operated (A, B, C), adrenalectomized (D, E, F) and adrenalectomized plus corticosterone (G, H, I) animals. Observe the decrease in dendritic branch points in all three cell types [granule cells located in the genu (A, D, G), granule cells with single primary dendrites (B, E, H) and granule cells with multiple primary dendrites (C, F, I)] with adrenalectomy compared with sham operation and adrenalectomy plus corticosterone.

length of the apical ($P > 0.25$) or basal dendrites ($P < 0.1$) in a 100-μm section (see Table 2). Moreover, concentric ring analysis showed no significant changes in the distribution of dendritic material with any treatment ($P > 0.1$ for all comparisons). Although no significant overall changes in cell body area ($P < 0.07$) or the density of excrescences ($P < 0.09$) were observed with treatment, a trend

Table 2

Morphological variable	Sham operation $n=4$	Adrenalectomy $n=4$	Adrenalectomy plus corticosterone $n=4$
Cell body area of CA3 pyramidal cells (μm^2)	341.5 ± 9.3	330.1 ± 6.5	357.6 ± 6.5
Number of dendritic branch points of CA3 pyramidal cell apical tree	9.0 ± 0.7	10.8 ± 1.1	11.8 ± 0.7
Number of dendritic branch points of CA3 pyramidal cell basal tree	9.5 ± 1.1	9.6 ± 0.7	10.6 ± 1.2
Length of apical dendrites within 100-μm section (μm)	1779.5 ± 239.1	1452.0 ± 91.3	1561.1 ± 70.3
Length of basal dendrites within 100-μm section (μm)	1431.7 ± 48.1	852.9 ± 165.1	1450.5 ± 184.0
Number of pyknotic cells in CA3 region/10^6 μm^2	0.69 ± 0.09	0.53 ± 0.08	0.69 ± 0.10
Number of apical excrescences/10 μm	11.9 ± 0.9	9.9 ± 0.5	11.8 ± 0.7
Cell body area of CA1 pyramidal cells (μm^2)	232.4 ± 4.9	223.9 ± 2.7	229.1 ± 6.6
Number of pyknotic cells in CA1 region/10^6 μm^2	0.91 ± 0.12	0.75 ± 0.10	0.92 ± 0.18

Values represent mean ± standard error. These data were analysed by one-way ANOVA with *post hoc* comparisons (see text). No significant differences were detected.

toward a decrease in both of these parameters was observed with adrenalectomy (Table 2).

Quantitative analysis of cell body area of CA1 pyramidal cells revealed no significant difference or trend toward a difference between adrenalectomized, sham operated and corticosterone replaced animals ($P > 0.25$; see Table 2).

Effects of glucocorticoid manipulations on hippocampal cell survival

Light microscopic examination of Nissl-stained sections revealed a few pyknotic cells in the dentate gyrus of sham operated animals at both three and seven days following surgery (Table 1). These degenerating profiles were almost always located deep in the granule cell layer or at the border between the granule cell layer and the hilus. Adrenalectomy resulted in dramatic increases in the number of pyknotic cells in the dentate gyrus by three days following surgery (Fig. 3). These degenerating cells were almost always located within the granule cell layer. Seven days following adrenalectomy, the mean number of pyknotic cells was greater than that observed at three days following adrenalectomy. Significant differences in the number of pyknotic cells were detected at this time ($P < 0.005$; see Table 1). In addition, regional differences in the distribution of these cells could be discerned; pyknotic cells, although located throughout the dentate gyrus, were more concentrated in the rostral aspect of the hippocampal formation. Furthermore, a greater number of these degenerating profiles was observed in the suprapyramidal blade of the dentate gyrus (Fig. 4). Within the suprapyramidal blade of the dentate gyrus, a concentration of pyknotic cells was seen in the lateral tip. The infrapyramidal blade of the dentate gyrus showed fewer pyknotic cells. At both three and seven days following adrenalectomy, the pyknotic cells in the dentate gyrus appeared to possess varying characteristics (Figs 3, 5). Some degenerating cells were larger than others and the larger population was more likely to show darker cytoplasm, more granules of condensed chromatin and processes which resembled dendrites than the smaller pyknotic cells (Figs 3, 5). These profiles are characteristic of cells in the earlier stages of degeneration.[5,7] Replacement of corticosterone in the drinking water to adrenalectomized animals prevented the appearance of large numbers of pyknotic cells in all regions of the dentate gyrus (Table 1).

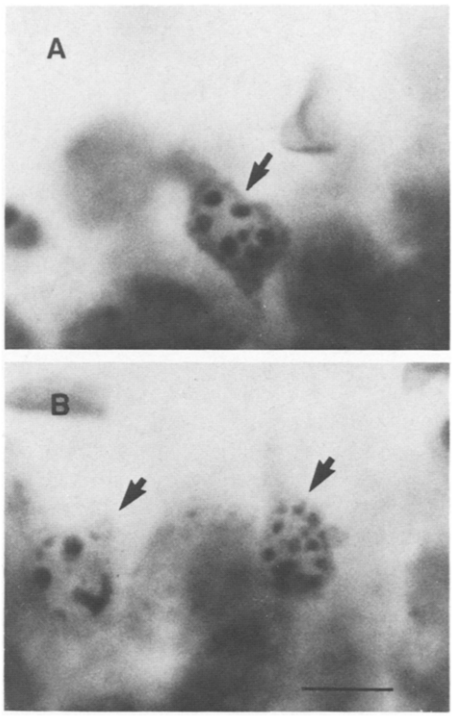

Fig. 3. Pyknotic cells (arrows) in the granule cell layer of the dentate gyrus of brains of adrenalectomized animals three days following surgery. Observe the condensed chromatin which is characteristic of degenerating cells in both A and B. Scale bar in B = 15 μm and applied to both frames.

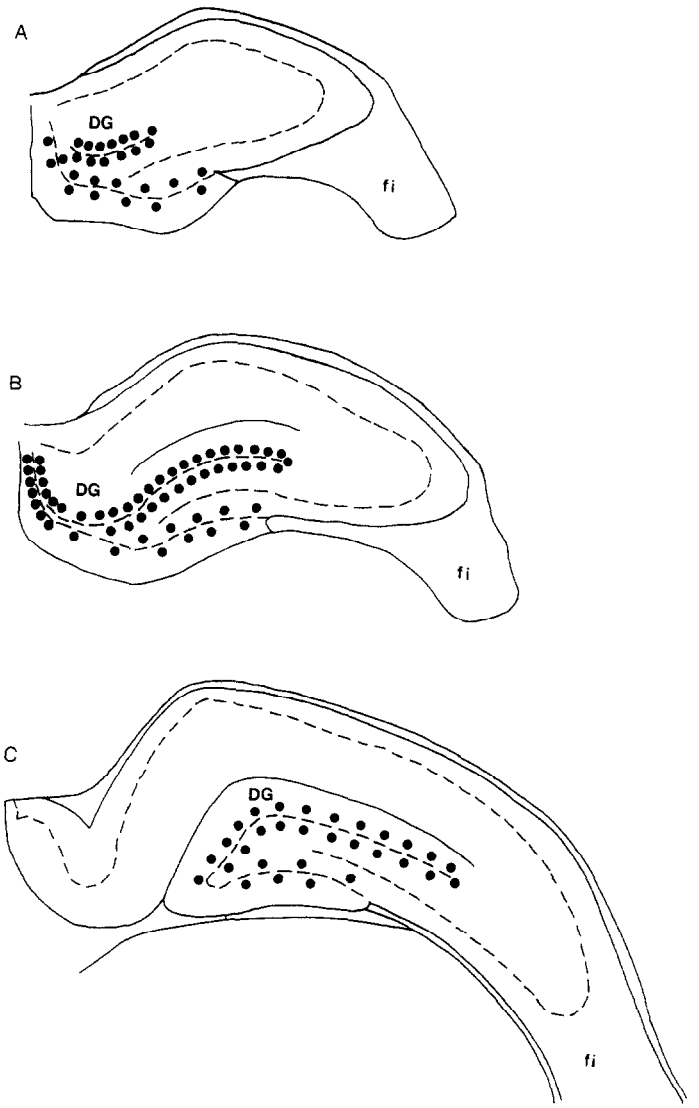

Fig. 4. Templates mapping the distribution of pyknotic cells at three different levels of the hippocampal formation from brains of adrenalectomized animals seven days following surgery. Each solid dot represents five pyknotic cells. Observe the concentration of degenerating cells in the suprapyramidal blade in A, B and C compared with the infrapyramidal blade. Templates A, B and C correspond to plates 27, 30 and 35 of Paxinos and Watson,[20] respectively. DG, dentate gyrus; fi, fimbria.

Light microscopic examination of the CA3 and CA1 regions of the hippocampus revealed very few pyknotic cells in all brains of sham operated animals (Table 2). Adrenalectomy resulted in no change in the number of degenerating cells in the pyramidal cell layers of the CA1 or CA3 regions at either three or seven days following surgery (see Table 2).

DISCUSSION

Glucocorticoid effects on granule cell morphology and survival

These results demonstrate that short-term manipulations of glucocorticoid levels have profound effects on granule cells of the dentate gyrus. Adrenalectomy caused significant degeneration, detectable by high numbers of pyknotic cells, throughout the dentate gyrus as rapidly as three days following surgery. By seven days following adrenalectomy, the number of pyknotic cells was even greater. Due to the thickness of the brain sections examined, it is likely that the pyknotic cell counts obtained in the present study underestimate the actual number of degenerating profiles present in a given area of the dentate gyrus granule cell layer. Moreover, cells in the earliest stages of degeneration cannot be detected with light microscopic analysis.[7] Despite these methodologic considerations, the number of pyknotic cells reported here is quite substantial. Developmental studies have shown that, even in neural regions where massive cell death is occurring, the number of pyknotic cells detectable at a given time point greatly

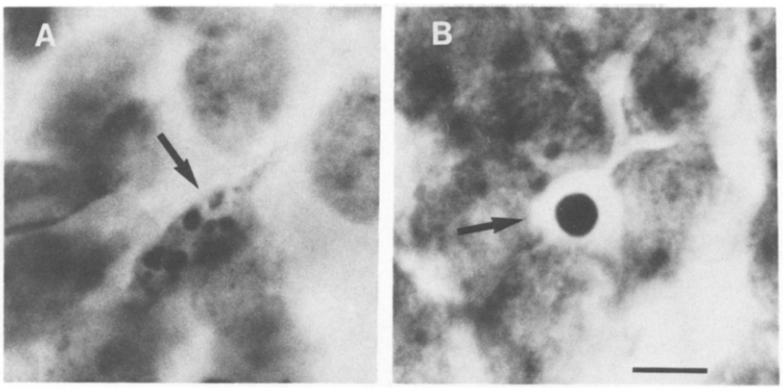

Fig. 5. Pyknotic cells (arrows) in the granule cell layer of the dentate gyrus of adrenalectomized animals seven days following surgery. Observe the condensed chromatin, a characteristic of degenerating cells, in both cases. Note also the neuron-like appearance of the pyknotic cell in A and the relatively late stage of degeneration, characterized by an absence of cytoplasm, of the pyknotic cells in B. Scale bar in B = 15 μm and applies to both frames.

underestimates the actual loss of cells.[7,12,25] The rapid course of both degeneration and removal of cellular debris (reported to occur, in some instances, as quickly as 1 and 7 h, respectively) probably accounts for this phenomenon.[12] Although degeneration and removal of cellular debris are thought to be less rapid in adult than developing animals,[7] it is likely that the large numbers of pyknotic cells observed in the adrenalectomized brains of this study reflect only a fraction of the actual cell loss.

Although it is conceivable that some of the pyknotic cells observed in the dentate gyrus following adrenalectomy represent the death of glia, there are several lines of evidence which suggest that adrenalectomy-induced cell death is primarily neuronal: (1) the degenerating cells we have observed were almost exclusively located in the granule cell layer of the dentate gyrus, a region which contains granule neurons and few, if any, glia;[15,22] (2) no pyknotic cells have been observed which stain for the astrocyte marker, glial fibrillary acidic protein (unpublished observation); and (3) the pyknotic cells often resembled neurons in that they were large and possessed dendrite-like processes. Collectively, these observations strongly support the contention that adrenalectomy results in cell death which is primarily neuronal.

The analysis of Golgi-impregnated cells revealed adrenalectomy-induced morphologic changes in granule cells which suggested that this neuronal population was degenerating. Adrenalectomy resulted in significant decreases in cell body area and numbers of dendritic branch points of Golgi-impregnated granule cells. Although these changes were observed in Golgi-impregnated cells after adrenalectomy, no obvious signs of morphologic abnormalities were observed in these cells. Since Nissl-stained tissue from the same brains revealed massive cell death in the dentate gyrus, it likely that the Golgi-impregnated neurons of brains of adrenalectomized animals examined in this study represented a population of granule cells which were in the earlier stages of atrophy, leading perhaps to degeneration. Indeed, many Nissl-stained pyknotic cells of the dentate gyrus of adrenalectomized animals possessed characteristics of the earlier stages of degeneration; stained cytoplasm, processes resembling dendrites and multiple granules of condensed chromatin.[5,7] It is also possible that our criteria for selecting Golgi-impregnated cells for morphologic analysis excluded those granule cells which are in the later stages of degeneration. For example, degenerating neurons may appear to be incompletely impregnated and would not be selected for analysis.

Although enormous variability exists within the dentate gyrus of adrenalectomized animals in the present study at both three and seven days post-surgery, all brains of adrenalectomized animals showed substantially more pyknosis than sham operated or corticosterone-replaced animals. Sloviter *et al.*[27] reported that approximately 35% of adrenalectomized animals did not show a total loss of the dentate gyrus over a three- to four-month interval. However, quantitative analysis of the dentate gyrus of adrenalectomized rats was not reported by these investigators. The results of the present study suggest that quantitative analysis of the dentate gyrus from adrenalectomized rats after a three- to four-month interval would reveal some cell loss, but probably to varying degrees in the entire treatment group. At least two possibilities could explain the variability observed in the brains of adrenalectomized animals in both the present report and the report of Sloviter *et al.*:[27] (1) accessory adrenal tissue could be present in those animals which exhibit less cell death; or (2) granule cells of some animals could be less dependent on glucocorticoids for their survival.

The appearance of large numbers of pyknotic cells as well as the decrease in cell body area and the number of dendritic branch points were prevented by providing corticosterone in the drinking water to adrenalectomized rats. These findings suggest that

other hormones which derive from the adrenal gland, such as catecholamines, are not necessary for the survival of dentate gyrus granule cells. Since granule cells express both Type 1 and Type 2 glucocorticoid receptors[10,21] it is possible that occupation of one or both of these receptor types is required for their survival. The extent to which Type 1 and Type 2 receptors mediate cell survival and the maintenance of normal morphology within the dentate gyrus remains to be determined.

Specificity of glucocorticoid effects in the hippocampal formation

Despite the presence of both Type 1 and Type 2 receptors in CA1 and CA3 pyramidal cells[10,21] the present report shows that these neurons do not require adrenal steroids for their survival. The lack of pyknotic cells in the pyramidal cell layer of the CA1 and CA3 regions of brains from adrenalectomized animals suggests that, within the hippocampal formation, massive degeneration is specific to the dentate gyrus. However, the observations of trends toward a decrease in cell body area and the density of excrescences in CA3 pyramidal cells of brains from adrenalectomized animals suggest that if later time points were examined, significant differences might be detectable in this region as well. Since excrescences of CA3 pyramidal cells are postsynaptic to granule cell mossy fibers,[16] it is likely that cell death of this afferent population would affect the postsynaptic elements. Further experimentation examining later time points will be necessary in order to confirm this possibility. Nonetheless, the absence of pyknotic cells in the CA3 region of brains from adrenalectomized animals suggests a fundamental difference between CA3 pyramidal cells and dentate gyrus granule cells in response to glucocorticoids. In addition to observing no pyknosis in the CA1 pyramidal cell layer of adrenalectomized rats, no change was observed in cell body area in this population as well. Sloviter *et al.*[27] have shown normal electrophysiologic responses in the CA1 region of adrenalectomized rats (with a three- to four-month interval between surgery and being killed). Collectively, this finding and those of the present report support the notion that, unlike dentate gyrus granule cells, pyramidal cells of the CA1 region do not require glucocorticoids for maintenance of normal morphology or physiology.

The results of the present study indicate that the dentate gyrus is unique among other regions of the hippocampal formation in that it undergoes naturally occurring cell death in adulthood. Unlike other regions of the hippocampal formation, in which pyknotic cells are seldom, if ever, observed, the dentate gyrus of sham operated animals shows a small but consistent number of pyknotic cells. Comparable numbers of pyknotic cells are also observed in the dentate gyrus of non-operated animals (unpublished observations). Recent studies performed in our laboratory suggest that naturally occurring cell death in the dentate gyrus is also under the control of glucocorticoids: (1) excess glucocorticoids administered to intact animals result in a significant decrease in dentate gyrus pyknosis (unpublished observations); and (2) natural fluctuations in the levels of circulating glucocorticoids with the diurnal rhythm[4] correspond to changes in the numbers of dying cells; during the dark phase, when glucocorticoid levels are high, numbers of pyknotic cells are lower whereas during the light phase, when glucocorticoid levels are low, numbers of pyknotic cells are higher (unpublished observations). The results of these experiments strongly suggest that glucocorticoids enhance cell survival in the intact dentate gyrus.

Granule cells of the adult dentate gyrus are also unique among hippocampal neurons in that the period of neurogenesis in this neuronal population extends throughout the postnatal period[1] and into adulthood in the rat.[2,6,14] Interestingly, the majority of pyknotic cells in the dentate gyrus of intact rats are observed at the sites of adult neurogenesis, the subgranular region[6] suggesting a relationship between cell birth and death in the dentate gyrus of adult intact rats.

Since developmental studies have shown that neurogenesis in certain brain regions, including the hippocampal formation, is affected by adrenalectomy[3,4] as well as glucocorticoid treatment,[3] it is possible that adrenalectomy in adulthood enhances cell turnover. That is, adrenalectomy may increase the rate of both cell division and cell death. Further experimentation will be necessary in order to determine the relationship between cell birth and death in the dentate gyrus as well as the extent to which neurogenesis in the adult dentate gyrus is under the control of glucocorticoids.

Acknowledgements—This research was supported by MH 41256 and by an equipment grant from the Richard Lounsbery Foundation. We thank Melissa Adaniel and Maryse V. Aubourg for their excellent technical assistance.

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(*Accepted* 20 *March* 1990)