Acta neuropath. (Berl.) 16, 95—102 (1970)

Quantitative Histological Studies on the Aged Human Brain

II. Senile Plaques and Neurofibrillary Tangles in Senile Dementia (with an Appendix on Their Occurrence in Cases of Carcinoma)

A. D. DAYAN

The National Hospitals for Nervous Diseases, Queen Square, London

Received February 7, 1970

Summary. Using a rigidly controlled quantitative histological technique the occurrence of senile plaques and argyrophilic tangles in the frontal cortex and Ammon's horn has been compared in "normal" controls and 40 selected cases of senile dementia.

There was a trend towards relatively larger numbers of neurofibrillary tangles in the dements than in the controls, and for these lesions to occur in particularly large numbers in Ammon's horn.

It was concluded that although a correlation may exist between the occurrence of senile dementia and the intensity of tangle formation, it is essential to take into account also the regional distribution in the brain of this and other types of lesions in relation to the specific clinical features of "senile dementia".

The brains of a further series of 20 patients with visceral carcinomatosis were examined for the premature occurrence of plaques ("Krebsdrusen"). None were found.

Zusammenfassung. Unter Verwendung einer streng kontrollierten quantitativen histologischen Technik wurde das Auftreten seniler Plaques und argyrophiler Fibrillen in Frontalrinde und Ammonshorn zwischen "normalen" Kontrollflächen und 40 ausgewählten Fällen von seniler Demenz verglichen. Es fand sich eine Tendenz zu relativ größerer Zahl Alzheimerscher Fibrillenveränderungen bei Dementen gegenüber den Kontrollgehirnen. Diese Veränderungen neigten zu besonders gehäuftem Auftreten im Ammonshorn.

Obwohl eine Korrelation zwischen dem Auftreten seniler Demenz und der Häufigkeit der Fibrillenveränderungen bestehen dürfte, wird vermutet, daß auch die regionale Verteilung dieser und anderer Läsionen im Gehirn mit den spezifischen klinischen Symptomen der „senilen Demenz" in Beziehung gesetzt werden müssen.

Die Gehirne einer weiteren Reihe von 20 Patienten mit Organcarcinomen wurden auf das vorzeitige Auftreten von Plaques („Krebsdrusen") untersucht. Solche wurden nicht gefunden.

Key-Words: Ageing Processes — Senile Plaques — Neurofibrillary Tangles — Quantitative Histological Study — Senile Dementia.

The nature and significance of "plaques and neurofibrillary tangles" in relation to varieties of dementia is uncertain, although both qualitative and quantitative attempts have been made to correlate these lesions with the occurrence and severity of so-called "senile dementia" and "Alzheimer's disease" (Simchowicz, 1910; Grünthal, 1927; Rothschild, 1937; Newton, 1948; von Braunmühl, 1957; Corsellis, 1962; and Blessed, Tomlinson and Roth, 1968).

The problems which have arisen in these studies have included achieving uniform diagnostic criteria and assessment of the degree of dementia of the patients during their lifetime, and, after death of measuring the frequency and intensity of the pathological changes studied. Since the realization (Simchowicz, 1910; Gellerstedt, 1933) that apparently similar lesions may be found in aged, normal

subjects, their role in cases of dementia has become even more obscure. Some workers, for example, Rothschild (1942) have invoked a variety of associated pathological and psychological factors to account for the dementia, whilst others have speculated that there might still be a relationship between these particular pathological changes and the psychiatric disorders, albeit a quantitative rather than a qualitative one (e.g. Corsellis, 1962; Blessed *et al.*, 1968).

This report describes a quantitative histological analysis of the intensity of plaques and neurofibrillary tangles in the brains of 40 cases of senile dementia, and comparison of the results with the changes found in 75 normal patients studied in a similar fashion (Dayan, 1970).

Material and Methods

The technical methods followed were those of Dayan (1970). In summary, blocks were taken at standard levels from coronal slices of the right middle frontal gyrus and Ammon's horn, and Glees-Marsland stained paraffin sections prepared under standardised conditions. The numbers of lesions were counted under direct vision, and, after allowing for the effects of section thickness and shrinkage, the results were corrected to a common base of lesions/cu.mm. of cortex of fixed brain. All values shown are the means of two counts. The standard deviation of individual results was very small and has not been shown for the sake of clarity.

The brains were obtained at autopsies on long-stay patients at two large psychiatric hospitals. On clinical grounds all the cases had been diagnosed by experienced psychiatrists as suffering from "senile dementia", although none had been subjected to detailed psychometric studies. Cases of dementia which showed any macroscopic evidence of vascular disease in the brain, or, of other possible causes of dementia or neoplasm anywhere in the body have been excluded from the analysis.

Results

The age composition of the series is shown in Table 1. 60 % of the series were females, but because the numbers of cases available was small, information for men and women has been pooled, and any possible effect due to sex has been ignored in analysing the results and comparing them with the normal controls.

The (corrected) numbers of plaques and tangles counted in Ammon's horn are shown in Fig. 1, and are compared with the corresponding values found in controls (Dayan, 1970). The separate values found in the frontal cortex and Ammon's horn are compared with each other in Fig. 2. Straight lines were fitted separately to the data for the plaque and tangle counts by the method of linear regression employing the least squares technique. The equations obtained (plaques $y = 0.83\,x + 52.1$; tangles $y = 0.94\,x + 273$), and their respective correlations statistically were significantly different ($p = 0.05$). This means that in the cases analysed there was a significantly greater number of neurofibrillary tangles in

Table 1. *Age composition of series of senile dements*

Age group (years)	No. of patients	
	Male	Female
60—69	8	10
70—79	4	10
79+	4	4

Total no. of cases = 40.

Ammon's horn as compared with the frontal cortex, whereas the intensity of plaques was similar at both sites as had been found previously in the normal controls.

Qualitative Observations

Apart from plaques and tangles, the histological lesions noted in the areas examined of these brains were banal, and comprised in all cases a variable loss of neurones, thickening and gliosis of the molecular layer of the cerebral cortex, and, loading of neurones and some glial cells with yellow lipofuscin pigments. Material with the staining properties of amyloid was not sought routinely. Such material was found however, in the small intra-cortical blood vessels fo 2 patients, one aged 56 years, and the other (aged 73 years) who had the highest plaque count recorded. It was sought in them because of the unusual thickening and hyalinisation of the walls of the vessels. Amyloid-like staining of plaques and tangles: magenta with periodic acid Schiff, salmon-pink with Congo red and giving an anomalous green colour in polarised light, was not sought in every specimen. In any case, these properties seemed relatively uncommon, particularly the latter

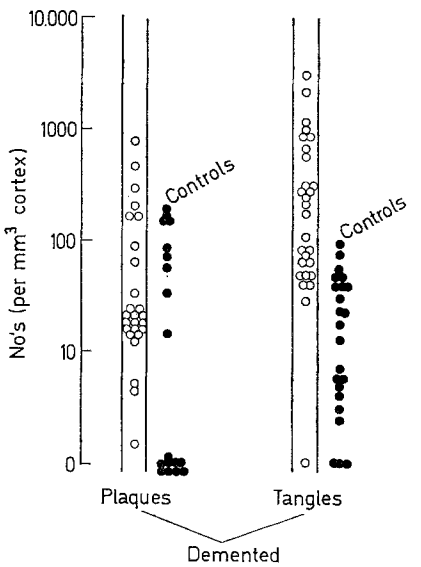

Fig. 1 The numbers of senile plaques and argyrophilic tangles found in Ammon's horn in cases of senile dementia are compared with the corresponding values found in controls

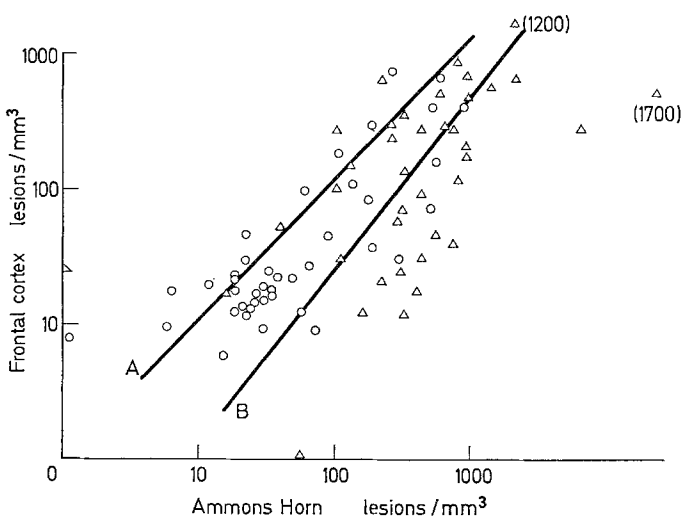

Fig. 2. Relationship between the numbers of plaques and tangles in the frontal cortex and Ammon's horn in cases of senile dementia. A is the independently calculated regression line for plaques. B is the line calculated for tangles (see text). ○ plaques; △ tangles

two, but this may have been due to the random nature and small size of the sample examined and the relative insensitivity of the techniques employed. This conclusion is supported by a comparison with the results of a survey employing more sensitive methods (Wright *et al.*, 1969); in which "amyloid" was found in the brains of 63 % of people more than 70 years old.

The occurrence of granulo-vacuolar degeneration (Simchowicz, 1911); in Ammon's horn was noted in the pyramidal neurones there in 40 % of cases. Small numbers of neurones appeared to be affected, mainly in Sommer's sector and the H2 region.

Discussion

The purpose of this work was to compare certain quantitative histological observations on the brains of cases of senile dementia with results obtained from a study of similar age-related changes in normal controls. The choice of features studied as well as areas examined was governed by the availability of data on control cases.

In essence, the results obtained appear to show that it is possible to differentiate cases of senile dementia from normal patients of similar age by means of the intensity of neurofibrillary tangle formation in the brain, particularly in Ammon's horn, as this appears to reach much higher levels in the demented patients.

This interpretation of the present results is as dependent on the method of selecting the demented patients, as on the techniques employed for counting and analysing the plaques and tangles. The subjects chosen had been in-patients at two psychiatric hospitals for periods ranging from several months to a few years. The diagnosis of "senile dementia" had been applied to them by several psychiatrists, who had almost certainly employed a variety of clinical criteria. Pathologically, the cases were further selected by the exclusion of all those with other organic lesions capable of producing psychiatric disorders.

It is not claimed that these patients are representative of the clinical entity of "senile dementia", only that they comprise a pathologically homogeneous group of cases of dementia occurring in later life. The manner of collection of the cases differentiates the series in some respects from those studied by other workers, who have usually accepted cases initially on purely clinical grounds (e.g. Simchowicz, 1910; Rothschild, 1937; Newton, 1948; Corsellis, 1962; Blessed *et al.*, 1968; Jamada and Mehraien, 1968), and subsequently have made attempts by internal analysis and deductive reasoning to allow for the effects of the cerebrovascular disease so commonly observed amongst the patients. By this artificial restriction of the type of disorder studied, it was hoped to delineate more clearly the possible significance of the pathological lesions, and subsequently, to examine these findings in terms of their clinical importance.

As shown in Fig. 2, no cases were entirely devoid of plaques and tangles at both sites examined, although each of two separate patients lacked either plaques or tangles in Ammon's horn, and another case had no tangles in the frontal cortex. This may be taken as some evidence for the homogeneity of the cases studied. Overall there was a very considerable scatter in the numbers of lesions present at both sites, and the figures obtained to some extent overlap with those found in control cases. There is no significant difference between the numbers of plaques

at either site when compared with the normals. However, there is a definite difference between the demented and normal patients if the numbers of neurofibrillary tangles at both sites are compared; and in the cases of dementia, there is a particularly strong trend towards an excess of tangles in the Ammon's horn. This achieves the conventional level of significance ($p = 0.05$), if the numbers of tangles in the frontal cortex and Ammon's horn in individual cases are compared statistically with controls of similar age, but a more detailed formal analysis has not been undertaken because it was not possible to match the demented and control patients sufficiently closely for them to be considered as samples of an homogeneous population. These findings cannot be compared directly with previous results because of the methodological differences in the selection of patients and the manner in which they have been studied.

Both the earliest quantitative histological studies of senile dementia (Simchowicz, 1911 and 1924) and the most recent (Blessed *et al.*, 1968) have demonstrated a suggestive correlation between the occurrence of dementia and the numbers of plaques present in the brain. The former showed also that the actual numbers of plaques present in different areas of the brain varied greatly, and even suggested a histological index for differentiating cases of senile dementia from examples of "Alzheimer's disease", based on the ratio between the numbers of plaques in the frontal and occipital cortex. It is difficult now to accept this distinction (Blessed *et al.*, 1968), although it is still supported by some, e.g. Wildi *et al.* (1964). Blessed *et al.* (1968) showed a highly significant correlation between an index of the numbers of plaques in the brain and psychometric measures of the degree of dementia and intellectual performance in elderly patients. By comparison with a group of similar patients suffering from functional disorders they showed also that age could not be correlated with increasing numbers of plaques.

These workers employed a "mean plaque count" obtained by averaging the numbers found in sections from 12 areas of the brain (excluding hippocampus). In view of the almost uniform report of considerable variation in the incidence of plaques in different areas of the brain, this global figure is of doubtful validity (e.g. Simchowicz, 1911; Grünthal, 1926; McMenemey, 1940; Goodman, 1953; Morel and Wildi, 1955; von Braunmühl, 1957; Sjögren and Sourander, 1961; Hirano and Zimmerman, 1962; Wildi *et al.*, 1964; Jamada and Mehraein, 1968); as it may well conceal extensive differences between the various areas. There are very few pathological processes which affect the brain so uniformly that a reliable quantitative parameter can be obtained by averaging values obtained from almost all areas of the cerebral cortex.

The occurrence within the clinical syndrome of senile dementia of cases with accentuation of particular symptoms and signs and the possibility of correlating these features with focal concentration of pathological lesions in the brain (e.g. Blessed *et al.*, 1968; Jamada and Mehraein, 1968; McDonald, 1969) provides further evidence that the changes in the brain are not randomly distributed, and should not be represented by a single parameter if a detailed analysis is to be attempted of the significance of the histological lesions studied. The finding that there appears to be a certain concordance between the incidence of both plaques tangles (e.g. Newton, 1948; von Braunmühl, 1957; present study) may account in part for the correlation found by Blessed *et al.* between the dementia score and

the "mean plaque count" in some of their cases. The present observation of an apparent correlation between the diagnosis of "Senile Dementia" and the numbers of neurofibrillary tangles at particular sites may also be specious for the same reasons.

In such patients it is not known to what extent the dementia is due to the overall loss of neurones so often noted, to qualitative or quantitative malfunctioning of neurones due to the presence of plaques and tangles which imply disordered neuronal ultrastructure (see review by Terry, 1969) and therefore metabolism, or to some combination of these and other factors. For these reasons at least, it would appear premature to attempt a general correlation of the clinical entity of "dementia" with only one or two of the many pathological processes which have affected the brains of these patients.

Appendix

In 1935 Neubürger and Rösch reported finding senile plaque-like structures ("Krebsdrusen") in 19 of 60 young patients with visceral carcinomas. Such lesions were seen in only 4 of 60 control cases of similar age (40—60 years). They concluded that a systemic metabolic reaction to the visceral neoplasms might somehow have stimulated the formation of the plaques.

In an attempt to repeat this observation, the brains of 20 unselected, consecutive patients who had died from visceral carcinomatosis, were examined by the same quantitative histological procedures as had been employed for the work described elsewhere in this report.

Details of the patients and their neoplasms are given in Table 2. All of them had widely disseminated metastases. Apart from some agonal confusion and

Table 2. *Patients dying from visceral carcinomas*

Case No.	Age (years)	Sex	Site of primary carcinoma	Cerebral metastases
1	47	M[a]	Lung	+[b]
2	51	F	Breast	+
3	55	F	Breast	+
4	55	F	Stomach	0
5	59	M	Colon	0
6	44	M	Lung	+
7	43	F	Breast	0
8	41	F	Stomach	+
9	56	M	Colon	0
10	46	F	Breast	0
11	52	M	Lung	+
12	59	F	Pharynx	0
13	49	F	Colon	0
14	44	M	Lung	0
15	57	F	Bladder	0
16	53	M	Stomach	+
17	57	M	Lung	+
18	57	M	Bladder	0
19	58	M	Colon	0
20	56	F	Kidney	+

[a] M = male; F = female; [b] + = present; 0 = absent.

thought disorders in a few of them, none of the patients had any record of mental disease on retrospective search of their records. The results of the histological survey were the demonstration of plaques: 4/cu.mm. in the frontal cortex of case 2, and 8/cu. mm. in the frontal cortex and Ammon's horn of case 4. No neurofibrillary tangles were found anywhere.

Thus, it was impossible to confirm the results of Neubürger and Rösch. The difference between the two series cannot readily be accounted for by differences in technique or errors of interpretation as the illustrations of Neubürger and Rösch (as they themselves noted), show typical appearing primitive and nucleated plaques, which do not look like Tinel's (1924) pseudoplaques or other staining artefacts; similarly, their cases appear to have been obtained from a general hospital and not from a psychiatric institution as only 3 of their cases (and none of the present series) had any previous history of mental disorders.

Although "Krebsdrusen" may not be a feature of unselected cases of carcinomatosis, it is still possible that a small number of unusual patients may bear them by analogy with the "carcinomatous neuropathies" which affect only a small proportion of cases. It would seem worthwhile examining a larger series of brains in an attempt to confirm the association between them and any particular type of neoplasm, and to see if any metabolic or other relationship could be demonstrated between the cortical and the visceral lesions.

Acknowledgements. I am grateful to Messrs. J. Mitchell and J. Mills for their skilled assistance, and for support from the British Empire Cancer Campaign for Research, and the Smith, Kline and French Foundation. I wish to thank Dr. J. Brierley for permitting me to examine some of his cases.

References

Blessed, G., Tomlinson, B. E., Roth, M.: The association between quantitative measures of dementia and of senile change in the cerebral grey matter of elderly subjects. Brit. J. Psychiat. **114**, 797—811 (1968).

Braunmühl, A. von: Alterserkrankungen des Zentralnervensystems In: Hdb. d. spez. Path. Anat. Bd. 13/1, ed. O. Lubarsch, F. Henke u. R. Rössle. Berlin-Göttingen-Heidelberg: Springer 1957.

Corsellis, J. A. N.: Mental Illness and the Ageing Brain. London 1962.

Dayan, A. D.: Quantitative histological studies on the aged human brain: I Senile plaques and neurofibrillary tangles in "normal" patients. Acta neuropath. (Berl.) **16**, 85—94 (1970).

Gellerstedt, N.: Zur Kenntnis der Hirnveränderungen bei der normalen Altersinvolution: Upsala Läk.-Fören. Förh. **38**, 194—408 (1933).

Goodman, L.: Alzheimer's disease: a clinicopathologic analysis of 23 cases with a theory of pathogenesis. J. nerv. ment. Dis. **117**, 97—130 (1953).

Hirano, A., Zimmerman, H. M.: Alzheimer's neurofibrillary changes. Arch. Neurol. (Chic.) **7**, 227—242 (1962).

Jamada, M., Mehraein, P.: Verteilungsmuster der senilen Veränderungen im Gehirn. Arch. Psychiat. Nervenkr. **211**, 304—324 (1968).

McDonald, C.: Clinical heterogeneity in senile dementia. Brit. J. Psychiat. **115**, 267—271 (1969).

McMenemey, W. H.: Alzheimer's disease: a report of six cases. J. Neurol. Psychiat. **3**, 211—239 (1940).

Morel, F., Wildi, E.: Contributions à la connaissance des différentes altérations cérébrales du grand âge. Arch. suisses Neurol. Psychiat. **76**, 174—223 (1955).

Neubürger, K., Rösch, A.: Über argentophile Ablagerungen im Gehirn bei Krebskranken. Virchows Arch. path Anat. **294**, 537—545 (1935).

Newton, R. D.: Identity of Alzheimer's disease and senile dementia and their relation to senility. J. ment. Sci. **94**, 225—249 (1948).

Rothschild, D.: Pathologic changes in senile psychoses and their psychologic significance. Amer. J. Psychiat. **93**, 757—788 (1937).

— Neuropathologic changes in arteriosclerotic psychoses and their psychiatric siginficance. Arch. Neurol. Psychiat. (Chic.) **48**, 417—437 (1942).

Sjögren, H., Sourander, P.: Histopathological studies in Alzheimer's disease. In: Proc. IVth Int. Congr. Neuropathol., vol. 3, pp. 319—324, ed. H. Jacob. Stuttgart 1962.

Simchowicz, T.: Sur la signification des plaques séniles et sur la formule sénile de l'écorce cérébrale. Rev. neurol. **1**, 221—227 (1911).

Terry, R. D.: Electron microscopic studies of Alzheimer's disease and of experimental neurofibrillary tangles. In: The Central Nervous System, pp. 213—224, ed. O. T. Bailey, and D. E. Smith. Baltimore 1969.

Tinel, J.: Les processus anatomo-pathologiques de la démence sénile. Rev. neurol. **11**, 23—49 (1924).

Wright, J. R., Calkins, E., Breen, W. J., Stolte, G., Schultz, R. T.: Relationship of amyloid to aging. Medicine (Baltimore) **48**, 39—60 (1969).

Wildi, E., Linder, A., Costoulas, G.: Étude statistique des altérations dégénératives cérébrales apparaissant au cours du viellissement. Psychiat. et Neurol. (Basel) **148**, 41—68 (1964).

The National Hospitals for Nervous Diseases
Department of Neuropathology
Queen Square, W.C. 1, London