RESULTS
D. RESULTS

1. ALTERATION OF GROWTH OF BODY AND BRAIN:

The degrees of deviations in the body weights and brain weights of the rats used for the electron microscopic (EM) studies presented in this thesis are presented below, so as to exemplify an indication of the magnitude of undernutritional effect, that prevailed behind the data.

a) **Body Weights**

i) **Growth in the normal (control) subjects:** Only male rats were used for the study. The results showed significant increase in the body weight with increasing age in the controls (Fig.R-1 and Table R-1). The percentage of increase was 25% between day 10 and 15, which increased to 92%, by 21 days, to 178% by 45 days and 131% by 150 days. At day 10 the mean body weight was 17.66 ± 1.63 gm, and by 150 days the mean body weight increased to 273.33 ± 4.08 gm.

ii) **Growth in the undernourished:** The undernourished group also showed significant ontogenetic increase in the body weight with increasing age (Fig.R-1 and Table R-1). However, at any given age as compared with the controls, the body weight of the undernourished was significantly less (range 38-55%) (Table R-1). The percentage of increase was 70% from day 10 to day 15, 50% between day 15 and 21 days, 179% by 45 days and 205% by 150 days.
<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Weight</th>
<th>(a) Control (n=6)</th>
<th>(b) Undernourished (n=6)</th>
<th>(c) Rehabilitated (n=5)</th>
<th>a vs b</th>
<th>a vs c</th>
<th>b vs c</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>Body</td>
<td>17.66 ± 1.630</td>
<td>7.830 ± 0.752</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Weight</td>
<td>1.08 ± 0.040</td>
<td>0.771 ± 0.045</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>15</td>
<td>Body</td>
<td>22.16 ± 2.040</td>
<td>13.330 ± 1.360</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Weight</td>
<td>1.27 ± 0.022</td>
<td>0.933 ± 0.052</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>21</td>
<td>Body</td>
<td>42.50 ± 2.730</td>
<td>20.00 ± 1.780</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Weight</td>
<td>1.58 ± 0.019</td>
<td>1.020 ± 0.025</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>45</td>
<td>Body</td>
<td>118.33 ± 6.83</td>
<td>55.80 ± 3.76</td>
<td>71.00 ± 4.183</td>
<td>***</td>
<td>***</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Weight</td>
<td>1.77 ± 0.020</td>
<td>1.236 ± 0.021</td>
<td>1.56 ± 0.038</td>
<td>***</td>
<td>***</td>
<td>***</td>
</tr>
<tr>
<td>Age (days)</td>
<td>Weight (gm)</td>
<td>(a) Control (n=6)</td>
<td>(b) Undernourished (n=6)</td>
<td>(c) Rehabilitated (n=5)</td>
<td>a vs b</td>
<td>a vs c</td>
<td>b vs c</td>
</tr>
<tr>
<td>-----------</td>
<td>-------------</td>
<td>------------------</td>
<td>--------------------------</td>
<td>-------------------------</td>
<td>-------</td>
<td>-------</td>
<td>-------</td>
</tr>
<tr>
<td>150</td>
<td>Body Weight</td>
<td>273.33 ± 4.080</td>
<td>170.000 ± 5.477</td>
<td>235.00 ± 13.690</td>
<td>***</td>
<td>***</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td>Brain Weight</td>
<td>2.02 ± 0.017</td>
<td>1.880 ± 0.021</td>
<td>1.96 ± 0.041</td>
<td>***</td>
<td>*</td>
<td>**</td>
</tr>
</tbody>
</table>

Students t-test: Significance levels

*** P < 0.001
** P < 0.005
* P < 0.010

n = number of animals
FIG. R-1.  BODY WEIGHTS IN GRAMS

C: CONTROL
UN: UNDERNOURISHED
Rh: REHABILITATED

*** P < 0.001 C vs UN
** P < 0.001 C vs Rh
* P < 0.001 UN vs Rh

AGE (Days)
iii) Growth in post-weaning rehabilitated: On rehabilitation significant improvement or gain in body weights was seen by both 45 days and 150 days compared to matched undernourished, but were still significantly less when compared with controls of the respective ages.

b) Brain Weights:

i) In normal group: The ontogenetic data on the brain weights of the controls showed an increase with increasing age (Fig.R-2 and Table R-1). Between day 15 and 21 the gain in brain weights was maximum (24%). At day 10 the brain weight was 1.08 ± 0.04 gm, which increased to a weight of 2.02 ± 0.01 gm by 150 days of age.

ii) In the undernourished group: In the undernourished, the ontogenetic increase in the brain weight was seen to be highest (53%) by 150 days. At day 10 it was 0.77 ± 0.04 gm and by 150 days it was 1.88 ± 0.02 gm.

Compared to the control at any respective age the undernourished had significantly less brain weight (Table R-1).

iii) In rehabilitated group: On rehabilitation the brain weight was seen to increase as compared with the age matched undernourished by 27% by day 45 and by 4% by 150 days, while, compared with the controls they were still significantly less.
FIG. R-2. ONTOGENY OF BRAIN WEIGHTS

UNDER NOURISHED ONTOGENY

CONTROL ONTOGENY

AGE (days)

BRAIN WEIGHT IN Gms

BRAIN WEIGHT IN Gms
# Table R-2(a)

**Percentage Differences in Body and Brain Weights**

<table>
<thead>
<tr>
<th>Weights</th>
<th>Deficit as Compared with Age Matched Controls</th>
<th>10 Days</th>
<th>15 Days</th>
<th>21 Days</th>
<th>45 Days</th>
<th>150 Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body Weight</td>
<td></td>
<td>55.66%</td>
<td>39.84%</td>
<td>52.94%</td>
<td>52.84%</td>
<td>37.80%</td>
</tr>
<tr>
<td>Brain Weight</td>
<td></td>
<td>28.60%</td>
<td>26.53%</td>
<td>35.44%</td>
<td>30.5%</td>
<td>7.0%</td>
</tr>
</tbody>
</table>

**Rehabilitated Compared with Controls**

<table>
<thead>
<tr>
<th>Weights</th>
<th>Deficit as Compared with Controls</th>
<th>40.0%</th>
<th>14%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body Weight</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brain Weight</td>
<td></td>
<td>11%</td>
<td>3%</td>
</tr>
</tbody>
</table>

**Rehabilitated Compared with Undernourished**

<table>
<thead>
<tr>
<th>Weights</th>
<th>Deficit as Compared with Undernourished</th>
<th>27%</th>
<th>38%</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body Weight</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Brain Weight</td>
<td></td>
<td>27%</td>
<td>4%</td>
</tr>
</tbody>
</table>
### TABLE: R - 2 (b)

**PERCENTAGE EXPRESSION OF BODY AND BRAIN WEIGHT GAIN WITH AGE**

<table>
<thead>
<tr>
<th>Nutritional status</th>
<th>10 - 15 Days</th>
<th>15 - 21 Days</th>
<th>21 - 45 Days</th>
<th>45 - 150 Days</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>CONTROL</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body</td>
<td>25.5%</td>
<td>91.8%</td>
<td>178.4%</td>
<td>131%</td>
</tr>
<tr>
<td>Brain</td>
<td>18%</td>
<td>24%</td>
<td>12%</td>
<td>14%</td>
</tr>
<tr>
<td><strong>UNDERNOURISHED</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body</td>
<td>70.33%</td>
<td>50%</td>
<td>179%</td>
<td>205%</td>
</tr>
<tr>
<td>Brain</td>
<td>20.7%</td>
<td>10%</td>
<td>20%</td>
<td>53%</td>
</tr>
</tbody>
</table>
In summary, the results of the ontogenetic changes in body weights and brain weights revealed that i) there was a significant decrease in the growth of both body and brain at all ages in the undernourished, and ii) post-weaning nutritional rehabilitation produced a substantial reduction in the development of the deficits in the weights compared to the group continued on undernutrition. Yet, there was a small and significant residual deficit persisting despite the rehabilitatory situation provided.

c) Serum protein level:

The mean serum protein levels in the control group was found to be $8.35 \pm 0.97 \text{ gm/dl}$, and in the undernourished $8.15 \pm 0.38 \text{ gm/dl}$ (Table R-3). The results showed that with the type of caloric undernutrition provided from the earliest of developing ages, no significant change in the serum protein levels occurred in the adults of the undernourished group.

2. CHANGES IN CINGULATE CORTEX MOLECULAR LAYER:

Data was collected on the molecular layer of cingulate cortex. Ages studied were 10, 15, 21, 45 and 150 days.
### TABLE: R - 3

**SERUM PROTEIN LEVELS**

**IN gms/dl**

<table>
<thead>
<tr>
<th>n</th>
<th>CONTROL</th>
<th>UNDERNOURISHED</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>9.40</td>
<td>8.42</td>
</tr>
<tr>
<td>2</td>
<td>7.35</td>
<td>7.65</td>
</tr>
<tr>
<td>3</td>
<td>9.10</td>
<td>8.62</td>
</tr>
<tr>
<td>4</td>
<td>8.60</td>
<td>8.13</td>
</tr>
<tr>
<td>5</td>
<td>7.30</td>
<td>7.93</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>n = 5</th>
<th>41.75</th>
<th>40.75</th>
<th>$\bar{x}$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>8.35</td>
<td>8.15</td>
<td>$\bar{x}$</td>
</tr>
</tbody>
</table>

| $\sigma_{n-1}$ | 0.97  | 0.38  |

Not Significant
a) **Thickness of molecular layer**: (Plate R-1)

i) **Control group**: The normal data on ontogeny revealed an increase in the thickness of molecular layer with increasing age from day 10 to 21 days, then a decrease at 45 days, followed by an increase again at 150 days (Fig.R-3 and Table R-4).

ii) **Undernourished group**: In the undernourished group there was a progressive increase in the thickness with increasing age from day 10 to 150 days, but the thickness of the layer was significantly less than that of the control up to 21 days, and more than that of the age matched controls at 45 and 150 days (Fig. R-3). There was no decrease noticed at 45 days, contrary to that seen in the control.

iii) **Rehabilitated group**: On rehabilitation surprisingly, the occurrence of increase in thickness of the molecular layer was prevented, and it had actually remained less than the control value (Fig.R-3). In other words, the occurrence of increase in thickness was prevented by rehabilitation, and the deficit of thickness continued to be like that prevailing before the time of rehabilitation (Fig.R-3).

iv) **Conclusion**: In summary the data revealed that chronic caloric undernutrition produced an initial lag in attainment of thickness of the molecular layer till 21 days, and a subsequent over shooting or excess relative to that of the age matched control. Rehabilitation prevented the overshooting effect that would have occurred under continuing undernutrition.
(THICKNESS OF MOLECULAR LAYER)

(ML)
# TABLE: R - 4

**THICKNESS OF MOLECULAR LAYER OF POSTERIOR CINGULATE CORTEX**

*Mean ± SD in μm*

<table>
<thead>
<tr>
<th>Age (days)</th>
<th>(a) Control (n=5)</th>
<th>(b) Undernourished (n=5)</th>
<th>(c) Rehabilitated (n=5)</th>
<th>a vs b</th>
<th>a vs c</th>
<th>b vs c</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>88.08 ± 0.42</td>
<td>53.43 ± 1.28</td>
<td>---</td>
<td>*****</td>
<td></td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>138.02 ± 11.91</td>
<td>115.03 ± 1.28</td>
<td>---</td>
<td></td>
<td>**</td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>151.23 ± 9.06</td>
<td>127.66 ± 12.83</td>
<td>---</td>
<td>****</td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td>136.29 ± 3.31</td>
<td>169.70 ± 5.95</td>
<td>119.96 ± 12.45</td>
<td>*****</td>
<td>**</td>
<td>*****</td>
</tr>
<tr>
<td>150</td>
<td>156.31 ± 3.81</td>
<td>176.17 ± 1.48</td>
<td>145.53 ± 0.94</td>
<td>*****</td>
<td>*****</td>
<td>*****</td>
</tr>
</tbody>
</table>

Student's t-test significance levels

- ***** P < 0.001
- **** P < 0.005
- ** P < 0.025

n = Number of rats
FIG. R.3 CINGULATE CORTEX MOLECULAR LAYER THICKNESS

C vs UN  ***** P < 0.001
C vs UN  **** P < 0.005
C vs UN  ** P < 0.025
UN vs Rh  fffff P < 0.001
C vs Rh  ***** P < 0.001
C vs Rh  **** P < 0.025

THICKNESS IN µm's

AGE (Days)
b) **Alterations in ontogeny of numerical density of synapses:** (Plate R - 2 to R - 7)

i) **Control group:** The density of synapses per unit area, in the molecular layer was seen to significantly increase with age from day 10 till 45 days, followed by a significant reduction by 150 days. The increase was 57% between 10 and 15 days, 89% between 15 and 21 days, and 30% between 21 and 45 days. This was followed by a significant decrease at 150 days by 56% (Fig. R-4 and Table R-5).

The data on the subtype of synapses revealed that at all ages studied the proportion (%) of flat type synapses was significantly more compared to the negative and positive types. Between day 15 and 45 days it was seen to be in the range of 72% to 77% and by 150 days it was seen to decrease to 54%. The proportion of positive synapses was also seen to increase with age. It was 8% at day 15 and increased to 28% by 150 days, while the negative type of synapses was seen to be lowest at 45 days, contributing only 5% of the synapses (Fig. R-5 and Table R-7).

ii) **Undernourished group:** At any given age, between day 10 and 45 days, the undernourished group showed a significantly less density of
TABLE : R - 5 (a)

DATA ON NUMERICAL DENSITY OF SYNAPSES IN THE MOLECULAR LAYER OF POSTERIOR CINGULATE CORTEX OF CONTROL AND EXPERIMENTAL RATS

Mean ± SD, per tissue area of 112 μm²

<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Control (n=6)</th>
<th>Undernourished (n=6)</th>
<th>Rehabilitated (n=5)</th>
<th>a vs b</th>
<th>a vs c</th>
<th>b vs c</th>
</tr>
</thead>
<tbody>
<tr>
<td>10</td>
<td>6.75 ± 0.62</td>
<td>2.91 ± 0.16</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>15</td>
<td>10.64 ± 0.37</td>
<td>6.25 ± 0.54</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>21</td>
<td>20.16 ± 1.63</td>
<td>9.52 ± 0.46</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>45</td>
<td>26.24 ± 0.75</td>
<td>12.69 ± 0.49</td>
<td>21.22 ± 2.81</td>
<td>***</td>
<td>**</td>
<td>***</td>
</tr>
<tr>
<td>150</td>
<td>11.46 ± 0.51</td>
<td>21.97 ± 1.65</td>
<td>12.87 ± 1.07</td>
<td>***</td>
<td>*</td>
<td>***</td>
</tr>
</tbody>
</table>

Students t-test Significance levels

*** P < 0.001
** P < 0.005
* P < 0.025

n = number of rats
### TABLE: R - 5 (b)

**PERCENTAGE EXPRESSION OF RATE OF ACQUISITION OF SYNAPSES IN THE MOLECULAR LAYER OF POSTERIOR CINGULATE CORTEX ONTOGENY**

<table>
<thead>
<tr>
<th>Nutritional Status</th>
<th>10 - 15 Days</th>
<th>15 - 21 Days</th>
<th>21 - 45 Days</th>
<th>45 - 150 Days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (Ad-lib)</td>
<td>57.62 %↑</td>
<td>89.47 %↑</td>
<td>30.15 %↑</td>
<td>56.32 %↓</td>
</tr>
<tr>
<td>Undernourished UN</td>
<td>114.70 %↑</td>
<td>52.32 %↑</td>
<td>33.29 %↑</td>
<td>73.12 %↑</td>
</tr>
<tr>
<td>Rehabilitated Rh</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>39.34 %↓</td>
</tr>
</tbody>
</table>

**Student's test**  ***   $P < 0.001$

**Significance levels**
synapses than those of the age matched controls. Contrasting this, later on, the density of synapses became significantly higher than that in the control group (Fig. R-4). At 150 days of age, the density synapses in the undernourished was between the values of normal groups of ages 21 and 45 days. Hence, whether due to a delay in the acquisition of synapses, or due to a genesis in excess, or due to a lack of dispersal in neuropil the undernourished group had a significantly elevated density of synapses at 150 days of age.

The density of synapses increased gradually but significantly. It was seen to increase by 115% between day 10 and 15 days. It increased by 52% between 15 and 21 days, by 33% between 21 and 45 days and by 73% between 45 and 150 days. There was no decrease in the density of synapses after 45 days of age, contrary to that seen in the controls (Table R-5).

Analysis of the subtypes of synapses revealed that the relative proportions of positive, flat and negative synapses in the undernourished group were similar to those in the normal group at 21 days of age. However, at the age below this (15 days) the proportions of negative and flat synapses were respectively higher and lower than corresponding values of control group; and these relativities were reversed by 150 days of age (i.e., the proportion of flat type of synapses were highest).
FIG. R-5. CINGULATE CORTEX

DIFFERENT TYPES OF SYNAPSES

CONTROL

△ FLAT
POSITIVE
○ NEGATIVE

SYNAPSES/102 µm² AREA

18
12
8
4

15 21 45 150

AGE (Days)

UNDERNOURISHED

△ FLAT
○ POSITIVE
○ NEGATIVE

NUMBER OF SYNAPSES/102 µm² AREA

30
24
18
12
6
0

10 15 21 45 150

AGE (Days)
FIG. R-6.
CINGULATE CORTEX  TYPES OF SYNAPSES

NEGATIVE TYPE SYNAPSES

FLAT OR STRAIGHT TYPE SYNAPSES

POSITIVE TYPE SYNAPSES

AGE IN DAYS
CINGULATE CORTEX

MOLECULAR LAYER (E-PTA STAINED SYNAPSES)

x 12,200 Mag.

10 DAYS

CONTROL

UNDERNOURISHED
Plate R-3

CINGULATE CORTEX
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
x 12,200 Mag.

15 DAYS

CONTROL

UNDERNOURISHED
Plate R-4

CINGULATE CORTEX
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
21 DAYS
x 12,200 Mag.

CONTROL

UNDERNOURISHED
CINGULATE CORTEX
MOLECULAR LAYER  (E-PTA STAINED SYNAPSES)
x 12,200 Mag.

45 DAYS

CONTROL

UNDERNOURISHED
CINGULATE CORTEX
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
x 12,200 Mag.

150 DAYS

CONTROL

UNDERNOURISHED
Plate R-7

CINGULATE CORTEX
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)

REHABILITATED - 45 DAYS

REHABILITATED - 150 DAYS
### TABLE : R - 6

**DIFFERENTIAL ANALYSIS OF SUB-TYPES OF SYNAPSES IN MOLECULAR LAYER OF POSTERIOR CINGULATE CORTEX**

*Mean ± SD, per tissue area of 112 μm²*

<table>
<thead>
<tr>
<th>Age (Days)</th>
<th>Types</th>
<th>(a) Control</th>
<th>(b) Undernourished</th>
<th>(c) Rehabilitated (a) vs (b)</th>
<th>(a) vs (c)</th>
<th>(b) vs (c)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>P</td>
<td>0.75 ± 0.550</td>
<td>0.40 ± 0.69</td>
<td>--</td>
<td>NS</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(20)</td>
<td>(10)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>6.75 ± 1.118</td>
<td>3.70 ± 1.70</td>
<td>--</td>
<td>****</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(20)</td>
<td>(10)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>1.90 ± 0.852</td>
<td>2.60 ± 0.96</td>
<td>--</td>
<td>NS</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(20)</td>
<td>(10)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>P</td>
<td>0.8 ± 0.91</td>
<td>0.45 ± 0.51</td>
<td>--</td>
<td>NS</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(20)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>13.50 ± 2.41</td>
<td>6.85 ± 1.78</td>
<td>--</td>
<td>****</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(20)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>3.10 ± 0.56</td>
<td>1.70 ± 1.03</td>
<td>--</td>
<td>****</td>
<td>--</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(20)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age (Days)</td>
<td>Types</td>
<td>(a) Control</td>
<td>(b) undernourished</td>
<td>(c) Rehabilitated (a) vs (b)</td>
<td>(a) vs (c)</td>
<td>(b) vs (c)</td>
</tr>
<tr>
<td>-----------</td>
<td>-------</td>
<td>-------------</td>
<td>---------------------</td>
<td>----------------------------</td>
<td>------------</td>
<td>------------</td>
</tr>
<tr>
<td>45</td>
<td>P</td>
<td>3.70 ± 1.76</td>
<td>1.00 ± 0.70</td>
<td>1.66 ± 1.11</td>
<td>**</td>
<td>***</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(5)</td>
<td>(15)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>13.30 ± 3.65</td>
<td>7.80 ± 1.30</td>
<td>14.53 ± 3.22</td>
<td>**</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(5)</td>
<td>(15)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>1.00 ± 1.24</td>
<td>2.40 ± 0.80</td>
<td>2.00 ± 1.73</td>
<td>†</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(5)</td>
<td>(15)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>150</td>
<td>P</td>
<td>3.10 ± 1.28</td>
<td>2.53 ± 1.24</td>
<td>1.83 ± 0.98</td>
<td>NS</td>
<td>**</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(15)</td>
<td>(18)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>F</td>
<td>5.90 ± 1.52</td>
<td>16.73 ± 1.53</td>
<td>8.66 ± 1.08</td>
<td>****</td>
<td>****</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(15)</td>
<td>(18)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>1.90 ± 0.73</td>
<td>0.66 ± 0.48</td>
<td>1.27 ± 0.57</td>
<td>****</td>
<td>*</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(10)</td>
<td>(15)</td>
<td>(18)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

P = Positive type  (n) = Number of EM fields  Students t-test Significance levels
N = Negative type  quantified in the rats
F = Flat type

**** P<0.001
*** P<0.005
** P<0.010
† P<0.050
* P<0.025
<table>
<thead>
<tr>
<th>Age (Days)</th>
<th>Types of Synapses</th>
<th>(a) Control</th>
<th>(b) Undernourished</th>
<th>(c) Rehabilitated</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>Positive</td>
<td>7.97%</td>
<td>5.97%</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>71.80%</td>
<td>55.22%</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>20.21%</td>
<td>38.80%</td>
<td>-</td>
</tr>
<tr>
<td>21</td>
<td>Positive</td>
<td>4.59%</td>
<td>5.00%</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>77.58%</td>
<td>76.11%</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>17.81%</td>
<td>18.88%</td>
<td>-</td>
</tr>
<tr>
<td>45</td>
<td>Positive</td>
<td>20.55%</td>
<td>8.92%</td>
<td>9.12%</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>73.88%</td>
<td>69.64%</td>
<td>79.87%</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>5.55%</td>
<td>21.42%</td>
<td>10.99%</td>
</tr>
<tr>
<td>150</td>
<td>Positive</td>
<td>28.44%</td>
<td>12.70%</td>
<td>15.56%</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>54.12%</td>
<td>83.98%</td>
<td>73.63%</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>17.43%</td>
<td>3.31%</td>
<td>10.79%</td>
</tr>
</tbody>
</table>
iii) **Rehabilitated group:** In the rehabilitated groups the densities of synapses were tending to be nearer to those of normal groups than to the values of undernourished groups (Fig. R-4 and Table R-5). At 45 days of age the number was lower than that of controls, but at 150 days of age it was nearly close to but higher than corresponding control (Fig. R-4). Interestingly, after introduction of rehabilitation, there was an increase in synapses at 45 days, but by 150 days there was a reduction by 39% (Table R-5). In this group also, the proportion of flat type of synapses was higher than in normal group. The density of positive synapses also was seen to increase between 45 and 150 days (Fig. R-5 and R-6). The rehabilitation seemed to improve the total density rather than alter the proportions of the subtypes of synapses.

3. **CHANGES IN NUMERICAL DENSITY OF SYNAPSES OF HIPPOCAMPAL (CA 1)–DENTATE MOLECULAR LAYER:** *(PLATE R-8 TO R-12)*

The data was collected from the molecular layer of the dorsal hippocampus (CA 1)–dentate region. Ages studied were 15, 21, 45 and 150 days.

i) **Control group:** The data revealed a gradual but significant increase in the density of synapses with increasing age. It was seen to increase by 49% from 15 to 21 days, and by 69% between 21 and 45 days, followed by an increase by 63% by 150 days (Table R-8a,b). It
was interesting to note that there was no decrease seen between 45 and 150 days (Fig. R-7), contrary to what was noticed in the ontogeny of cingulate cortex.

Analysis of the subtype of synapses revealed more proportions of flat type of synapses at any given age. At 150 days of age it was seen to contribute 81% of the total, in contrast to only 54% in the cingulate cortex (Table R-9 a,b).

The proportion of positive synapses was seen to increase with age from 2% at day 15 to 15% by 150 days (Fig. R-9). The proportion of negative type synapses was seen to be lowest at 150 days of age.

ii) Undernourished group: When compared with the controls, at all ages between 21 and 150 days, the undernourished was always seen to have significantly less number of synapses. This was in contrast to the cingulate cortex wherein it was significantly higher at 150 days compared to the controls (Fig. R-7).

There was an increase of only 3% seen in the density of synapses between 15 and 21 days, while 71% increase was seen in the number between 21 and 45 days of age followed by 9% increase by 150 days (Table R-8 a,b).

Analysis of the subtypes of synapses showed the prominence of the flat
<table>
<thead>
<tr>
<th>Age (days)</th>
<th>(a) Control (n = 6)</th>
<th>(b) Undernourished (n=6)</th>
<th>(c) Rehabilitated (n=5)</th>
<th>a vs b</th>
<th>a vs c</th>
<th>b vs c</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>5.63 ± 0.33</td>
<td>6.33 ± 0.15</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>21</td>
<td>8.41 ± 0.34</td>
<td>6.51 ± 0.97</td>
<td>-</td>
<td>***</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>45</td>
<td>14.20 ± 0.48</td>
<td>11.15 ± 0.45</td>
<td>11.58 ± 0.51</td>
<td>***</td>
<td>***</td>
<td>NS</td>
</tr>
<tr>
<td>150</td>
<td>23.18 ± 0.34</td>
<td>12.18 ± 0.56</td>
<td>10.88 ± 0.57</td>
<td>***</td>
<td>***</td>
<td>**</td>
</tr>
</tbody>
</table>

Students t-test  

** P < 0.005

Significance level

NS = Not significant

n = number of rats
**TABLE : R - 8 (b)**

PERCENTAGE EXPRESSION OF RATE OF ACQUISITION OF SYNAPSES IN MOLLEcular LAYER OF CA1 HIPPOCAMPUS - DENTATE REGION ONTOGENY

<table>
<thead>
<tr>
<th>Nutritional status</th>
<th>15-21 days</th>
<th>21-45 days</th>
<th>45-150 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control (ad-lib) C</td>
<td>*** 49.37%↑</td>
<td>*** 68.84%↑</td>
<td>*** 63.23%↑</td>
</tr>
<tr>
<td>Under-nourished UN</td>
<td>NS 2.84%↑</td>
<td>*** 71.27%↑</td>
<td>9.23%↑</td>
</tr>
<tr>
<td>Rehabilitated Rh</td>
<td>-</td>
<td>-</td>
<td>NS 6.04%↓</td>
</tr>
</tbody>
</table>

Student's t-test *** $P \leq 0.001$

Significance levels

NS - Not Significant
**FIG. R-7.** HIPPOCAMPUS (CA1) & DENTATE REGION

Molecular layer

C vs UN \[*** P < 0.001\]

C vs Rh

UN vs Rh \[SS P < 0.005\]

UN vs Rh NS Not significant

C : CONTROL

UN: UNDERNOURISHED

Rh: REHABILITATED

**NUMBER OF SYNAPSES/59 \(\mu\text{m}^2\) area**

**AGE (Days)**
type of synapses followed by the negative and the positive. 71% to 83% of the synapses were of the flat type. The positive and negative types were at their highest at 150 days: 14% and 15% respectively. The lowest density of positive type was seen at 21 days (3%) while the lowest density of negative type was seen at 15 days (7%) (Table R-9 a,b). On the whole, the relative proportions of the subtypes of synapses were not markedly different in undernourished from normal, the difference was mainly in the total density of synapses between the two groups.

iii) Rehabilitated group: On rehabilitation no recovery from the early effects of undernutrition was seen. The density had not increased between 45 days and 150 days despite the rehabilitatory nutrition, while in the normal group there was an increase by 63% during this period. The densities in the rehabilitated group remained at the same levels as in the undernourished group (Table 8 a,b).

Analysis of the subtypes revealed a trend of increase in the proportion of positive synapses by 150 days. This was 35% compared to 16% at 45 days. In the undernourished group, the corresponding values were only 14% and 8%, respectively. This seemed to be the only improvement due to rehabilitation.
Plate R-8

HIPPOCAMPUS-DENTATE
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
x 20,000 Mag.

15 DAYS

CONTROL

UNDERNOURISHED
Plate R-9

HIPPOCAMPUS-DENTATE
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
21 DAYS

CONTROL

UNDERNOURISHED
Plate R-10

HIPPOCAMPUS-DENTATE
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
x 20,000 Mag.

45 DAYS

CONTROL

UNDERNOURISHED
Plate R-11

HIPPOCAMPUS–DENTATE
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
× 20,000 Mag.

150 DAYS

CONTROL

UNDERNOURISHED
Plate R-12
HIPPOCAMPUS-DENTATE
MOLECULAR LAYER (E-PTA STAINED SYNAPSES)
x 20,000 Mag.

REHABILITATED - 45 DAYS

REHABILITATED - 150 DAYS
FIG. R-8
HIPPOCAMPUS (CA1) TYPES OF SYNAPSES & DENTATE

NEGATIVE TYPE SYNAPSES

FLAT OR STRAIGHT TYPE SYNAPSES

POSITIVE TYPE SYNAPSES

NUMBER OF SYNAPSES/59 μm² AREA

15  21  45  150
FIG. R-9. HIPPOCAMPUS (CA1) MOLECULAR LAYER & DENTATE

CONTROL

\( \Delta \) : FLAT

\( \bullet \) : POSITIVE

\( \circ \) : NEGATIVE

UNDERNOURISHED

NUMBER OF SYNAPSES / 59 \( \mu \text{m}^2 \) AREA

CONTROL ONTOGENY

UNDERNOURISHED ONTOGENY

AGE (Days)

AGE (Days)
### TABLE: R-9 (a)

Differential analysis of sub-types of synapses in molecular layer of (CA1) – dentate region of hippocampus

Mean ± SD per tissue area of 59 μm²

<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Types</th>
<th>(a) Control</th>
<th>(b) Undernourished</th>
<th>(c) Rehabilitated</th>
<th>a vs b</th>
<th>a vs c</th>
<th>b vs c</th>
</tr>
</thead>
<tbody>
<tr>
<td>P</td>
<td></td>
<td>0.10 ± 0.31</td>
<td>0.60 ± 0.54</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=10)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>F</td>
<td>5.10 ± 0.73</td>
<td>5.00 ± 0.70</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=10)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>0.80 ± 0.78</td>
<td>0.40 ± 0.54</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=10)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>P</td>
<td></td>
<td>0.80 ± 0.78</td>
<td>0.20 ± 0.44</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=10)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>21</td>
<td>F</td>
<td>5.80 ± 2.39</td>
<td>5.20 ± 0.44</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=10)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>2.30 ± 2.05</td>
<td>1.00 ± 0.70</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=10)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>P</td>
<td></td>
<td>1.60 ± 0.54</td>
<td>0.60 ± 0.89</td>
<td>1.60 ± 0.54</td>
<td>NS</td>
<td>NS</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=5)</td>
<td>(n=5)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>45</td>
<td>F</td>
<td>7.80 ± 1.78</td>
<td>5.80 ± 0.83</td>
<td>6.40 ± 1.14</td>
<td></td>
<td>*</td>
<td>NS</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=5)</td>
<td>(n=5)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>1.40 ± 0.89</td>
<td>0.80 ± 0.83</td>
<td>2.20 ± 0.83</td>
<td></td>
<td>NS</td>
<td>**</td>
</tr>
<tr>
<td></td>
<td></td>
<td>(n=5)</td>
<td>(n=5)</td>
<td>(n=5)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Age (days)</td>
<td>Types</td>
<td>(a) Control</td>
<td>(b) Undernourished</td>
<td>(c) Rehabilitated</td>
<td>a vs b</td>
<td>a vs c</td>
<td>b vs c</td>
</tr>
<tr>
<td>-----------</td>
<td>-------</td>
<td>-------------</td>
<td>--------------------</td>
<td>-------------------</td>
<td>--------</td>
<td>--------</td>
<td>--------</td>
</tr>
<tr>
<td>P</td>
<td></td>
<td>3.00 ± 1.05 (n=10)</td>
<td>1.50 ± 0.52 (n=10)</td>
<td>3.50 ± 1.47 (n=10)</td>
<td>***</td>
<td>NS</td>
<td>***</td>
</tr>
<tr>
<td>150</td>
<td>F</td>
<td>16.30 ± 2.11 (n=10)</td>
<td>7.80 ± 1.13 (n=10)</td>
<td>4.80 ± 1.47 (n=10)</td>
<td>***</td>
<td>NS</td>
<td>***</td>
</tr>
<tr>
<td>N</td>
<td></td>
<td>0.70 ± 0.67 (n=10)</td>
<td>1.60 ± 0.96 (n=10)</td>
<td>1.80 ± 1.31 (n=10)</td>
<td>**</td>
<td>*</td>
<td>NS</td>
</tr>
</tbody>
</table>

P Postitive type, F Flat type N Negative type

Student's test * P < 0.050

Significance levels ** P < 0.025

*** P < 0.001

n = number of EM fields quantified in the rats
**TABLE : R - 9(b)**

PROPORTION OF POSITIVE, FLAT AND NEGATIVE SYNAPSES TO TOTAL OF ALL SYNAPSES (PERCENTAGE)

**HIPPOCAMPUS - DENTATE**

<table>
<thead>
<tr>
<th>Age (days)</th>
<th>Types of Synapses</th>
<th>(a) Control</th>
<th>(b) Undernourished</th>
<th>(c) Rehabilitated</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>Positive</td>
<td>1.66 %</td>
<td>10 %</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>85 %</td>
<td>83 %</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>13.63 %</td>
<td>6.6 %</td>
<td>-</td>
</tr>
<tr>
<td>21</td>
<td>Positive</td>
<td>8.98 %</td>
<td>3.12 %</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>65.16 %</td>
<td>81.25 %</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>25.84 %</td>
<td>15.62 %</td>
<td>-</td>
</tr>
<tr>
<td>45</td>
<td>Positive</td>
<td>14.81 %</td>
<td>8.33 %</td>
<td>15.68 %</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>72.22 %</td>
<td>80.55 %</td>
<td>62.74 %</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>12.96 %</td>
<td>11.11 %</td>
<td>21.56 %</td>
</tr>
<tr>
<td>150</td>
<td>Positive</td>
<td>15 %</td>
<td>13.76 %</td>
<td>34.65 %</td>
</tr>
<tr>
<td></td>
<td>Flat</td>
<td>81.5 %</td>
<td>71.55 %</td>
<td>47.52 %</td>
</tr>
<tr>
<td></td>
<td>Negative</td>
<td>3.5 %</td>
<td>14.67 %</td>
<td>17.82 %</td>
</tr>
</tbody>
</table>
4. CATECHOLAMINERGIC AXONS: STUDY OF INTERVARICOSITY DISTANCE:

The varicosities on noradrenergic axons represent the synaptic regions (presynaptic) and are known to contain transmitter stores in vesicles. Measurement of the intervaricosity distance along fluorescent preparation of axon gives information of alterations in density of synapses formed by an axon. Such data were collected in both the control and undernourished animals, in both peripheral nervous system and cerebral cortex, to assess any effects that might be caused by chronic undernutrition on the catecholaminergic axons and synapse formation.

a) Iris: sympathetic postganglionic adrenergic axon fibres: Peripheral Nervous System (PNS):

The study was carried out on the sympathetic innervation of iris in 4 to 5 months old rats (Fig. R-11a, b).

In the controls the mean intervaricosity distance was observed to be $6.94 \pm 0.98 \mu m$, and in the chronically undernourished it was $7.06 \mu m \pm 0.98 \mu m$ (Table R-10 a,b). There was no statistically significant difference between the two values, thereby indicating that the density of synapses contributed by an axon fiber is not altered by chronic undernutrition.
TABLE: R - 10 (a)

MEAN INTER-VARICOSITY DISTANCE IN $\mu$m's

<table>
<thead>
<tr>
<th>Number</th>
<th>Mean (Control)</th>
<th>Number</th>
<th>Mean (Undernourished)</th>
</tr>
</thead>
<tbody>
<tr>
<td>n=10</td>
<td></td>
<td>n=11</td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>6.34</td>
<td>1</td>
<td>7.94</td>
</tr>
<tr>
<td>2</td>
<td>6.84</td>
<td>2</td>
<td>6.40</td>
</tr>
<tr>
<td>3</td>
<td>8.05</td>
<td>3</td>
<td>6.11</td>
</tr>
<tr>
<td>4</td>
<td>7.31</td>
<td>4</td>
<td>7.79</td>
</tr>
<tr>
<td>5</td>
<td>6.92</td>
<td>5</td>
<td>5.02</td>
</tr>
<tr>
<td>6</td>
<td>5.09</td>
<td>6</td>
<td>6.05</td>
</tr>
<tr>
<td>7</td>
<td>6.61</td>
<td>7</td>
<td>7.26</td>
</tr>
<tr>
<td>8</td>
<td>5.96</td>
<td>8</td>
<td>6.14</td>
</tr>
<tr>
<td>9</td>
<td>8.71</td>
<td>9</td>
<td>6.32</td>
</tr>
<tr>
<td>10</td>
<td>7.60</td>
<td>10</td>
<td>8.23</td>
</tr>
<tr>
<td></td>
<td></td>
<td>11</td>
<td>7.06</td>
</tr>
</tbody>
</table>

**Mean** 6.94  **SD ± 1.04**  **Mean** 7.06  **SD ± 0.98**
TABLE :  \( R - 10 \) (b)

<table>
<thead>
<tr>
<th>Number of Animals</th>
<th>Nutrition Type</th>
<th>Intervaricosity Distance ( \mu m )</th>
<th>Number of varicosities in 20 ( \mu m ) length</th>
</tr>
</thead>
<tbody>
<tr>
<td>( n )</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>AD–LIB Control</td>
<td>6.9484 ± 1.04</td>
<td>3.083 ± 0.47</td>
</tr>
<tr>
<td>11</td>
<td>Under Nourished</td>
<td>7.0694 ± 0.98</td>
<td>3.1367 ± 0.43</td>
</tr>
</tbody>
</table>

Difference between the Groups  
\[ t = 0.443246 \quad p = < 0.500^* \]  
\[ t = 0.027303 \quad p = < 0.500^* \]

\(^*\) (In-significant)
RAT IRIS
AGE: ADULT
BODY WEIGHT: 225 grams
CATEGORY: CONTROL

CAMERA LUCIDA TRACING

DISTANCE = 72.87 MICRONS 8 8 0 0
DISTANCE = 75.29 MICRONS 9 8 0 0
DISTANCE = 71.66 MICRONS 10 8 0 0
DISTANCE = 70.79 MICRONS 11 5 0 0
DISTANCE = 48.63 MICRONS 12 11 0 0
DISTANCE = 82.64 MICRONS 13 9 0 0
DISTANCE = 57.21 MICRONS 14 5 0 0
DISTANCE = 65.26 MICRONS 15 7 0 0
DISTANCE = 61.71 MICRONS 16 7 0 0
DISTANCE = 49.99 MICRONS 17 13 0 0
DISTANCE = 29.77 MICRONS 18 10 0 0
DISTANCE = 72.73 MICRONS 19 9 0 0
DISTANCE = 62.56 MICRONS 20 8 0 0
DISTANCE = 50.82 MICRONS 21 8 0 0
DISTANCE = 44.93 MICRONS 22 8 0 0
DISTANCE = 69.07 MICRONS 23 9 0 0
DISTANCE = 54.90 MICRONS 24 2 0 0
DISTANCE = 68.31 MICRONS 25 18 2
DISTANCE = 50.64 MICRONS 26 5 0 0
DISTANCE = 50.23 MICRONS 27 1 0 0
b) **Frontal cortex : Central adrenergic axon fibers : Central Nervous System (CNS)**

The ages studied were 5, 15, 21 and 50/60 days (Fig. R-12a,b; 13a,b; 14a,b; 15a,b).

The body weights of these animals is shown in Table R-11.

In the ontogeny of control group the intervaricosity distance at 5 days of age was more than at the three ages (Table R-11). It was 6.48 ± 0.44 µm at days 5 and 5.14 ± 0.30 µm at 21 days. Thus, there seemed to be a small increase in the density of varicosities up to 21 days which stabilized more or less at that level.

The undernourished group significantly differed from control group at 5 days of age only (Table R-11 and Fig. R-10). The distance was less in the undernourished group than in control, i.e., the density of varicosities per unit length of axon was more at that age than in control. At later ages, this difference disappeared, but the intervaricosity distances in the undernourished group were usually less than that of age matched control subjects, though not significant statistically.

In summary, the above results indicate the chronic caloric undernutrition may not alter much the adrenergic intervaricosity distance, i.e., the density of adrenergic synaptic contacts per unit length of axon fiber, in both the PNS but not so in CNS.
### TABLE: R - 11

**A** - Body weights (gm) of control and experimental rats used for histofluorescence study of catecholaminergic axon terminals  
**B** - Intervericosity distance (um's) of catecholaminergic axon terminals in frontal cortex of rat

<table>
<thead>
<tr>
<th>Age days</th>
<th>Body weight (gm)</th>
<th>Mean ± SD</th>
<th>a vs b</th>
<th>Intervericosity distance (um's)</th>
<th>Mean ± SD</th>
<th>a vs b</th>
</tr>
</thead>
<tbody>
<tr>
<td>(a)</td>
<td>(b)</td>
<td></td>
<td></td>
<td>(a)</td>
<td>(b)</td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>Undernourished</td>
<td></td>
<td></td>
<td>Control</td>
<td>Undernourished</td>
<td></td>
</tr>
<tr>
<td>(n=6)</td>
<td>(n=6)</td>
<td></td>
<td></td>
<td>(n=6)</td>
<td>(n=6)</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>11.00 ± 0.89</td>
<td>7.00 ± 0.89</td>
<td>***</td>
<td>6.48 ± 0.44</td>
<td>5.22 ± 0.26</td>
<td>***</td>
</tr>
<tr>
<td>15</td>
<td>24.16 ± 2.40</td>
<td>13.33 ± 2.65</td>
<td>***</td>
<td>5.73 ± 0.47</td>
<td>5.66 ± 0.69</td>
<td>NS</td>
</tr>
<tr>
<td>21</td>
<td>43.33 ± 7.52</td>
<td>15.50 ± 2.07</td>
<td>***</td>
<td>5.14 ± 0.30</td>
<td>$ 5.39 ± 0.42</td>
<td>NS</td>
</tr>
<tr>
<td>50/60</td>
<td>122.14 ± 16.03</td>
<td>51.85 ± 15.82</td>
<td>***</td>
<td>5.87 ± 0.78</td>
<td>5.18 ± 0.79</td>
<td>NS</td>
</tr>
</tbody>
</table>

*** P < 0.001     NS = Not significant     $ = Significant (P < 0.001) compare to the value at 5 days
FIG. FRONTAL CORTEX (GLYOXYLIC ACID INDUCED HISTO-R-10 FLUORESCENCE)

Mean intervaricosity distance (μms) of catecholaminergic axons

C CONTROL
UN UNDERNOURISHED
** P < 0.001
NS NOT SIGNIFICANT

AGE IN DAYS

<table>
<thead>
<tr>
<th>AGE IN DAYS</th>
<th>CONTROL (C)</th>
<th>UNDERNOURISHED (UN)</th>
</tr>
</thead>
<tbody>
<tr>
<td>15</td>
<td>n=6</td>
<td>n=6</td>
</tr>
<tr>
<td>21</td>
<td>n=6 NS</td>
<td>n=6</td>
</tr>
<tr>
<td>50/60</td>
<td>n=6 NS</td>
<td>n=1</td>
</tr>
</tbody>
</table>
Fig. R - 12a

catecholaminergic axons

histofluorescence

rat frontal cortex

age: 5 days

body weight: 11 grams

category: control

camera lucida tracing

computer printout

Distance = 45.46 microns 5 0 8 0
Distance = 63.91 microns 5 4 0 0
Distance = 46.32 microns 7 4 0 0
Distance = 63.69 microns 8 7 0 0
Distance = 56.93 microns 9 5 0 0
Distance = 26.86 microns 10 6 0 0
Distance = 17.02 microns 11 8 0 0
Distance = 39.63 microns 12 5 0 0
Distance = 31.65 microns 13 4 0 0
Distance = 53.72 microns 14 6 0 0
Distance = 57.45 microns 15 10 0 0
Distance = 42.2 microns 16 4 0 0
Distance = 16.65 microns 17 6 0 0
Distance = 38.77 microns 18 0 0
Distance = 52.07 microns 19 6 0 0
Distance = 25.54 microns 20 2 0 0
Σ: 102

A2 6
D2 1
RAT FRONTAL CORTEX
ACE: 5 days
BODY WEIGHT: 8 grams
CATEGORY: UNDERNOURISHED

CATECHOLAMINERGIC AXONS
HISTOFLUORESCENCE

Fig. R - 12b

CAMERA LUCIDA TRACING

COMPUTER PRINTOUT

DISTANCE = 32.14 MICRONS
DISTANCE = 59.4 MICRONS
DISTANCE = 61.05 MICRONS
DISTANCE = 58.12 MICRONS
DISTANCE = 55.77 MICRONS
DISTANCE = 33.39 MICRONS
DISTANCE = 66.05 MICRONS
DISTANCE = 33.29 MICRONS
DISTANCE = 49.04 MICRONS
DISTANCE = 58.0 MICRONS
DISTANCE = 112.82 MICRONS
DISTANCE = 27.38 MICRONS
DISTANCE = 27.63 MICRONS
DISTANCE = 73.68 MICRONS
DISTANCE = 53.77 MICRONS
RAT FRONTAL CORTEX
AGE: 21 days
BODY WEIGHT: 45 grams
CATEGORY: CONTROL

CATECHOLAMINERGIC AXONS
HISTOFLUORESCENCE

CAMERA LUCIDA TRACING

COMPUTER PRINTOUT

DISTANCE = 51.58 MICRONS 414.00
DISTANCE = 38.18 MICRONS 59.00
DISTANCE = 38.76 MICRONS 69.00
DISTANCE = 92.66 MICRONS 69.00
DISTANCE = 44.28 MICRONS 99.00
DISTANCE = 44.86 MICRONS 109.00
DISTANCE = 45.29 MICRONS 119.00
DISTANCE = 45.3 MICRONS 119.00
DISTANCE = 44.86 MICRONS 139.00
DISTANCE = 36.52 MICRONS 149.00
DISTANCE = 54.92 MICRONS 159.00
DISTANCE = 44.04 MICRONS 159.00
DISTANCE = 55.0 MICRONS 1113.00
DISTANCE = 41.52 MICRONS 90.00
DISTANCE = 44.65 MICRONS 01.00
Σ2 139
A2 9
A2 0
RAT FRONTAL CORTEX
AGE: 21 days
BODY WEIGHT: 15 grams
CATEGORY: UNDERNOURISHED

Fig. R - 14b
CATECHOLAMINERGIC AXONS
HISTOFUOORESCENCE

CAMERA LUCIDA TRACING

COMPUTER PRINTOUT

DISTANCE = 61.86 MICRONS 5 9 0 0
DISTANCE = 49.69 MICRONS 5 6 0 0
DISTANCE = 30.98 MICRONS 7 8 0 0
DISTANCE = 40.11 MICRONS 8 7 0 0
DISTANCE = 50.78 MICRONS 9 8 0 0
DISTANCE = 42.99 MICRONS 16 7 0 0
DISTANCE = 46.92 MICRONS 11 7 0 0
DISTANCE = 39.55 MICRONS 12 7 0 0
DISTANCE = 46.58 MICRONS 13 7 0 0
DISTANCE = 40.71 MICRONS 14 9 0 0
DISTANCE = 38.18 MICRONS 15 7 0 0
DISTANCE = 40.22 MICRONS 16 7 0 0
DISTANCE = 38.48 MICRONS 21 1 0 0
DISTANCE = 46.93 MICRONS 21 7 0 0
DISTANCE = 41.93 MICRONS 21 1 0 0
Z2 1 1 1
A2 7
D2 0
Fig. R-15a
CATECHOLAMINERGIC AXONS
HISTOFLUORESCENCE

RAT FRONTAL CORTEX
AGE: 50 days
BODY WEIGHT: 100 grams
CATEGORY: CONTROL

CAMERA LUCIDA TRACING

DISTANCE = 43.25 MICRONS
DISTANCE = 31.22 MICRONS
DISTANCE = 61.29 MICRONS
DISTANCE = 24.76 MICRONS
DISTANCE = 20.35 MICRONS
DISTANCE = 42.89 MICRONS
DISTANCE = 29.25 MICRONS
DISTANCE = 45.3 MICRONS
DISTANCE = 47.76 MICRONS
DISTANCE = 58.512 MICRONS
DISTANCE = 38.15 MICRONS
DISTANCE = 46.36 MICRONS
DISTANCE = 68.49 MICRONS
DISTANCE = 58.43 MICRONS
DISTANCE = 32.38 MICRONS

COMPUTER PRINTOUT
Fig. R-15b
CATECHOLAMINERGIC AXONS
HISTOFLUORESCENCE

RAT FRONTAL CORTEX
AGE: 50 days
BODY WEIGHT: 68 grams
CATEGORY: UNDERNOURISHED

CAMERA LUCIDA TRACING

COMPUTER PRINTOUT

DISTANCE = 115.56 MICRONS
DISTANCE = 52.1 MICRONS
DISTANCE = 105.45 MICRONS
DISTANCE = 38.53 MICRONS
DISTANCE = 28.65 MICRONS
DISTANCE = 100.41 MICRONS
DISTANCE = 51.03 MICRONS
DISTANCE = 72.22 MICRONS
DISTANCE = 49.26 MICRONS
DISTANCE = 33.94 MICRONS
DISTANCE = 79.9 MICRONS
DISTANCE = 36.12 MICRONS
DISTANCE = 31.43 MICRONS
DISTANCE = 85.37 MICRONS
DISTANCE = 32.9 MICRONS