Histidine Decarboxylase and DOPA Decarboxylase in the Stomach of the Developing Rat

A system of enterochromaffin-like cells[1,2] in the parietal cell region of the stomach is the major storage site of gastric histamine[3–5] in the rat. These cells, which normally are devoid of monoamines, have the capacity to produce and store, for instance, 5-hydroxytryptamine (5HT) and dopamine but only after administration of their respective precursors[1,2]. The cellular localization of histamine has been established by a recently developed histochemical technique in which freeze-dried tissue sections are exposed to *o*-phthalaldehyde (OPT) vapor, whereby histamine is converted into a fluorophor, which can be detected in the fluorescence microscope[3,4,6].

The gastric mucosa of the rat is known to contain high activities of both the specific L-histidine decarboxylase (EC 4.1.1.22) and the non-specific aromatic L-amino acid decarboxylase, also referred to as 3,4-dihydroxyphenylalanine (DOPA) decarboxylase (EC 4.1.1.26). Recently it was shown that the major portion of gastric histidine decarboxylase in the rat is contained in the system of histamine-storing enterochromaffin-like cells[5,7]. Gastric DOPA decarboxylase, on the other hand, is found both in these cells[1,2,7] and in the 5HT-storing enterochromaffin cells of the antral region[7,8]. While the DOPA decarboxylase activity of the enterochromaffin cells much exceeds that of the histamine-storing enterochromaffin-like cells[7], there is no evidence that histidine decarboxylase occurs in the enterochromaffin cell system. Although both histidine decarboxylase and DOPA decarboxylase may be found in mast cells[9,10] and possibly also in some unidentified exocrine gland cells, the bulk of these 2 enzymes in the rat stomach is found in amine-storing cells of the enterochromaffin and enterochromaffin-like type[7].

The development of gastric histidine decarboxylase and DOPA decarboxylase in the fetal and postnatal rat was studied in order to obtain further information on the cytochemical properties and physiological significance of the enterochromaffin-like cell system. The entire stomach wall (from fetal and new-born rats pooled stomach walls) were homogenized in 0.02 *M* phosphate buffer, pH 6.5 to a final concentration of 100 mg tissue/ml. Aliquots of the homogenate, usually corresponding to 25 mg tissue (wet weight), were taken for enzyme assays. Histidine decarboxylase and DOPA decarboxylase were both determined by incubating the extract with 1-C^{14}-labeled substrates in a total volume of 0.5 ml at pH 6.9 under nitrogen for 30 min at 37 °C (Table I). The enzyme activity was estimated by determination of the $C^{14}O_2$ produced[11]. All assays were made in duplicate and the enzyme activities were expressed as nmoles CO_2 formed/mg and h. Corrections were made for heated enzyme blanks.

High activities of gastric histidine decarboxylase could be demonstrated in the fetal rat 17–19 days after mating (Table II). The activity declined to non-measurable levels just before birth and stayed low until 6–8 days after birth. At this stage the histidine decarboxylase activity started to increase but adult values were not reached until approximately 1 month after birth (Figure). Adult values of DOPA decarboxylase could be demonstrated in the fetal rat stomach 19 days after mating. The enzyme activity remained fairly constant during subsequent fetal and postnatal stages (Figure).

The development of amino acid decarboxylases in the rat stomach appears to be related to the maturation of the enterochromaffin and enterochromaffin-like cell systems. Enterochromaffin cells can be demonstrated in the digestive tract quite early in the fetal development and their capacity to produce and store 5HT is fully established well before birth[13]. This seems to correspond to the early appearance of gastric DOPA decarboxylase.

The high histidine decarboxylase activity of the stomach during fetal development is coincident with a generally high histamine-forming capacity in tissues of the fetal rat[14] and confirms earlier observations by KAHLSON et al.[15].

Table I. Incubation conditions

Incubation ingredients	Histidine decarboxylase assay	DOPA decarboxylase assay
DL-DOPA-1-C^{14}; 0.2 mc/m*M*	–	8×10^{-4} *M*
L-Histidine-1-C^{14}; 10 mc/mM	4×10^{-5} *M*	–
Pyridoxal-5-phosphate	10^{-5} *M*	10^{-5} *M*
Glutathione	5×10^{-4} *M*	5×10^{-4} *M*
Aminoguanidine	10^{-4} *M*	–
Phosphate buffer	0.04 *M*, pH 6.9	0.04 *M*, pH 6.9

Table II. Histidine decarboxylase and DOPA decarboxylase in the fetal rat stomach[a]

Days after mating	Histidine decarboxylase activity, nmoles CO_2/mg/h	DOPA decarboxylase activity, nmoles CO_2/mg/h
17	55.8×10^{-4}	–
18	42.6×10^{-4}	–
19	15.0×10^{-4}	17.8×10^{-2}
20	1.4×10^{-4}	17.8×10^{-2}
21	0.2×10^{-4}	–
22 – at birth	0	7×10^{-2}

[a] Stomachs were pooled from all embryos of the litter, cut open, blotted on filter paper and weighed. Special care was taken in dissection to avoid contamination of the stomach with hepatic and pancreatic tissue. The results given are from duplicate determinations on 1 litter at each stage.

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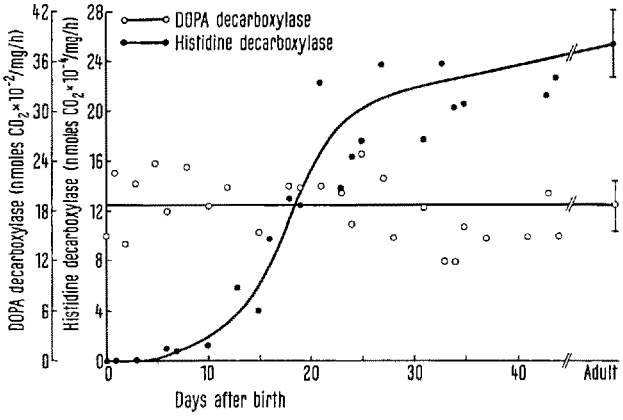
The decrease in gastric histidine decarboxylase shortly before birth follows the general decline of the activity of this enzyme in other tissues. It is unlikely that the gastric histidine decarboxylase of the fetal rat is related to a specific cell system. The potent enzyme activity may perhaps reflect the high mitotic activity at this stage[14]. In spite of the high histidine decarboxylase activity of the stomach of the fetal rat, its histamine content is quite low[4]. The enterochromaffin-like cells have the capacity to produce and store monoamines already at the time of birth[13]. Histamine, however, cannot be demonstrated histochemically in these cells until about 6–8 days after birth and adult levels are observed one month later[4]. The increase in gastric histamine seems to be correlated with the change in activity of histidine decarboxylase, indicating a local synthesis of the amine within the enterochromaffin-like cells. Apparently the cytochemical properties of the enterochromaffin-like cell system are not fully established until approximately 1 month after birth.

It was noted that shortly after the onset of the increase in gastric histidine decarboxylase the young rat started supplementing milk with adult rat food (16–18 days after birth). At the stage when the adult enzyme activity was attained, adult food habits were established. The maturation process of the enterochromaffin-like cells also seems to coincide with the establishment of normal hydrochloric acid secretion[16].

Gastric histidine decarboxylase in the adult rat is an adaptive enzyme and can be activated by a number of physiological and pharmacological stimuli[7,17,18]. Prolonged starvation causes a marked reduction of the gastric histidine decarboxylase activity whereas injection of gastrin to starved adult rats results in a 10:20-fold activation. In the young rat (14–17 days old), however, neither starvation nor injection of gastrin affected the enzyme activity (Table III). This may indicate that during this period gastric histidine decarboxylase is already maximally activated and that the regulation mechanism which relates the enzyme activity to food intake and starvation has not yet been established. After 18–19 days of age, starvation caused a marked decrease in gastric histidine decarboxylase, which could be counteracted by the injection of gastrin[19].



Activities of histidine decarboxylase and DOPA decarboxylase in the stomach of the developing rat. All animals were killed between 09.00 and 10.00 to ensure a uniform state of feeding. The whole stomach wall was taken for enzyme assays. Adult values are the mean of 7 determinations (vertical bars give standard error of the mean); all other values are the mean of 2–4 determinations. Each determination was made in duplicate. The curves were fitted visually.

Table III. Effect of starvation and gastrin injection on gastric histidine decarboxylase in young and adult rats

Age in days	Histidine decarboxylase activity nmoles $CO_2 \times 10^{-4}$/mg/h Starvation[a]	Gastrin[b]
14	5.0 ± 1.4 (3)	4.0 (2)
15	9.8 (2)	9.2 (2)
16	10.4 ± 5.4 (4)	9.2 ± 3 (4)
17	20.0 ± 5.6 (4)	28.0 ± 2.6 (4)
18	8.0 ± 4.8 (5)	21.0 (2)
19	3.8 ± 1.4 (5)	15.4 (2)
20	1.4 ± 0.4 (5)	15.0 ± 4 (4)
25	0.6 ± 0.4 (3)	10.6 ± 0.6 (3)
30	1.6 ± 1.4 (5)	14.0 ± 2.8 (3)
Adult	1.6 ± 1.2 (4)	18.0 ± 2 (6)

[a] All young rats were starved for 20–36 h. Adult rats were starved for 36 h. The whole stomach wall was taken for enzyme assays. [b] Synthetic human gastrin[12] was obtained through the courtesy of the American Gastroenterological Society. Gastrin was injected s.c. in a dose of 15 µg/kg. The rats were sacrificed 90 min after injection.

Zusammenfassung. Im Magen der fötalen Ratte erreicht die Dopadecarboxylase-Aktivität 19 Tage nach der Paarung Werte, die den Aktivitätswerten des erwachsenen Tieres entsprechen. Die Aktivität bleibt auch während der weiteren Entwicklung hoch. Die Histidindecarboxylase des fötalen Rattenmagens ist während der embryonalen Entwicklung hoch, aber die Aktivitätswerte fallen bei der Geburt zu nicht messbaren Werten ab. Ungefähr 8 Tage post partum steigt die Histidindecarboxylase des Magens wieder an und erreicht einen Monat nach der Geburt Werte, die den Aktivitätswerten des erwachsenen Tieres entsprechen. In der jungen Ratte wird die Magenhistidindecarboxylase weder durch Gastrininjektion noch durch Hungern beeinträchtigt. Vom 18. Tag nach der Geburt an kann die Enzymaktivität durch Hungern verringert und durch Gastrin wieder erhöht werden.

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