The Influence of Some Biogenic Amines and Cyclic N-2-O-Dibutyryl-Adenosine-3',5'-Monophosphate on Glycogen Content in Rat Brain Slices

The most obvious system for biochemical investigation of cyclic-3',5'-adenosinemonophosphate (CAMP) action in brain is that of control of glycogen metabolism, because it is only in this area that mechanism of CAMP action is understood with a considerable degree of biochemical precision. While adrenaline stimulates the adenyl cyclase activity of brain extracts, particularly of the cerebellum[1], no effect of the other biogenic amines on glycogen metabolism of the brain under normal conditions has been clearly demonstrated[2,3]. On the other hand, the activity of glycogen phosphorylase is modified by catecholamines and by CAMP, the metabolism of which is also affected by catecholamines[4]. Intraperitoneal injection of 3,4-dehydroxy-1-phenylalanine together with an amine oxidase inhibitor (β-phenylisopropylhydrazine) into mice results in a diminution of brain glycogen with an increase in catecholamines, while when the concentration of the latter is decreased, the level of glycogen returns to its normal value[5]. Although the stimulatory effect of catecholamines on adenyl cyclase of cerebellar homogenate could not be reproducted by others[6], KAKIUCHI and RALL[7,8] have shown that, in cerebellar slices, catecholamines stimulated the accumulation of CAMP. Recently, it was shown that the increased accumulation of CAMP was not specific for norepinephrine and dopamine; histamine and serotonin also raised CAMP levels[7-9]. On the other hand, the glycogenolytic effect of epinephrine[10] and serotonin[11] in rat brain slices was demonstrated.

The results obtained (Table) show that epinephrine, norepinephrine, histamine, serotonin and db-CAMP, as well as dopamine and CAMP, but to lesser degree, decreased glycogen concentration in rat brain slices (cortex, caudate, thalamus). It is of interest to point out the glycogenolytic effect of CAMP which was small but distinct in the brain tissue ($p < 0.05$) while in diaphragm CAMP itself did not change the concentration of glycogen[15].

The data obtained indicate that CAMP might be a 'second messenger' mediator in glycogenolytic influence of some biogenic amines in central nervous system and that epinephrine, norepinephrine, dopamine, as well as serotonin and histamine activate glycogen phosphorylase in the brain slices via adenyl cyclase-cyclic AMP system.

Résumé. On a montré que l'adrénaline, la noradrénaline, l'histamine, la sérotonine, le dérivé dibutyrique du 3',5'-AMP cyclique et aussi, bien qu'à un moindre degré, la dopamine et le 3',5'-AMP cyclique réduisent in vitro le glycogène du cerveau des rats.

B. B. MRŠULJA

Institute of Biochemistry,
Faculty of Medicine,
YU-11001 Belgrade 7
(Yugoslavia), 17 January 1972.

The effect of epinephrine, norepinephrine, dopamine, histamine, serotonin, CAMP and db-CAMP on glycogen concentration in rat brain slices

Treatment of the tissue	Brain slices		
	Cortex	Caudate	Thalamus
1. Controls	28.3±1.3	45.0±1.3	15.3±1.0
2. Epinephrine (10^{-4} μM/ml)	15.5±1.7[a]	21.0±1.3[a]	8.0±0.9[a]
3. Norepinephrine (10^{-4} μM/ml)	19.6±1.2[a]	25.3±1.3[a]	7.0±1.0[a]
4. Dopamine (10^{-4} μM/ml)	21.4±1.0[b]	27.3±2.1[b]	10.0±0.9[b]
5. Histamine (10^{-4} μM/ml)	17.5±1.0[a]	24.1±2.2[a]	6.8±1.0[a]
6. Serotonin (10^{-4} μM/ml)	16.1±1.3[a]	25.1±1.2[a]	8.5±1.0[a]
7. CAMP (10^{-3} μM/ml)	20.8±1.3[b]	28.4±1.4[b]	8.2±1.4[b]
8. db-CAMP (10^{-3} μM/ml)	11.4±1.1[a]	20.0±1.1[a]	5.0±1.1[a]

The amount of glycogen is expressed in ml/100 mg of tissue. The numbers indicate the mean value (M) of 5 experiments ±S.E.M. [a] $p < 0.01$ in comparison with the controls. [b] $p < 0.05$ in comparison with the controls.

As it was shown that biogenic amines accumulate the CAMP content in the tissues, it was of interest to know whether they have any influence on glycogen content in rat brain tissue. Taking into account the possible difference in penetration characteristics between CAMP and N-2-O-dibutyryl-adenosine-3',5'-monophosphate (db-CAMP), it was also of interest to compare the effects of these 2 substances on glycogen concentration in different brain slices of the rat.

The experiments were carried out on adult male Wistar rats. Brain slices were prepared according to the method already described[12] and were allowed 10 min in saline[12] at 37 °C. Epinephrine (10^{-4} μM/ml), norepinephrine (10^{-4} μM/ml), dopamine (10^{-4} μM/ml), histamine (10^{-4} μM/ml) and seratonin (10^{-4} μM/ml), as well as CAMP (10^{-3} μM/ml) and db-CAMP (10^{-3} μM/ml) were added at the beginning of the incubation, and immediately after 10 min from the brain slices glycogen was extracted[13] and estimated[14].

[1] L. M. KLEINER, Y. M. CHI, S. L. FREIDBERG, T. W. RALL and E. W. SUTHERLAND, J. biol. Chem. *237*, 1239 (1962).
[2] S. E. KERR and M. GHANTUS, J. biol. Chem. *116*, 9 (1936).
[3] R. V. COXON, E. C. GORDON-SMITH and J. R. HENDERSON, Biochem. J. *97*, 776 (1965).
[4] E. W. SUTHERLAND, T. W. RALL and T. MENON, J. biol. Chem. *48*, 237 (1962).
[5] Y. KAKIMOTO, T. NAKAJIMA, M. TEKESADA and T. SATO, J. Neurochem. *11*, 431 (1963).
[6] W. P. BURKARD and K. F. GEY, Helv. phys. Acta *26* CR, 197 (1968).
[7] S. KAKIUCHI and T. W. RALL, Molec. Pharmac. *4*, 367 (1968).
[8] S. KAKIUCHI and T. W. RALL, Molec. Pharmac. *4*, 379 (1968).
[9] M. HUANG, H. SHIMIZU and J. DALY, Molec. Pharmac. *7*, 155 (1971).
[10] B. B. MRŠULJA and L. M. RAKIĆ, J. Neurochem. *17*, 821 (1970).
[11] B. B. MRŠULJA, Iug. physiol. pharmac. Acta *7*, 131 (1971).
[12] R. RODNIGHT and H. MCILWAIN, Biochem. J. *57*, 649 (1954).
[13] F. N. LEBARON, Biochem. J. *61*, 80 (1955).
[14] R. MONTGOMERY, Arch. Biochem. *67*, 378 (1957).
[15] V. M. VARAGIĆ, M. ŽUGIĆ and B. B, MRŠULJA, Experientia *28*, 305 (1972).