autosomes(12,13). The mechanism controlling the appearance of the metacentrics is not clear. Ruddle suggests that it may be due to terminal union of 2 acrocentrics(14) but other mechanisms cannot be excluded. However, the differences observed in number and morphology of metacentric chromosomes provides convenient markers for cell lines with otherwise indistinguishable acrocentric autosomes.

Summary. An established line of canine kidney cells (MDCK-USD) of male origin was shown to contain 3 metacentric chromosomes and possess a modal number of 81. It grew rapidly in Eagle's MEM, displayed contact inhibition and was susceptible to vaccinia, VSV and influenza (PR8) viruses. Two additional MDCK lines, the parent line and another derivative from other laboratories, were compared to the USD line and could be distinguished on the basis of modal number and morphology of the metacentric chromosomes observed in each line.

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Normal Acid-Base Composition of Cerebrospinal Fluid in Infants And Children.* (31294)

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Much recent work has focused attention upon the acid-base composition of the cerebrospinal fluid (CSF) and its relation to the acid-base composition of the blood. This relationship is of general interest in an understanding of the functional nature of the blood-CSF barrier and of specific interest in elucidation of the mechanism for the chemical control of pulmonary ventilation(1). To date all studies of blood acid-base relationships have been carried out on either experimental animals or on adult human subjects. In connection with studies in progress in this laboratory on the CSF acid-base status of infants and children(2), the need for precise definition of normal CSF acid-base composition of patients in these age groups became apparent. The present paper presents such data.

Methods. Patients. Two groups of patients were studied: (a) a group of 11 infants of low birth weight and (b) a group of 13 children. Clinical data on the infants are shown in Table I. Birth weight varied from 1050 to 2100 g. All were clinically normal, having been continuously observed since birth in the Premature Nursery of the Babies Hospital. Because of the lack of a precise range

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TABLE I. Data on Normal Infants.

Patient	Age (days)	Sex	Race	Plasma pCO_2 (mm Hg)	Plasma pH	Plasma $[HCO_3^-]$ (mEq/l plasma w.)	CSF pCO_2 (mm Hg)	CSF pH	CSF $[HCO_3^-]$ (mEq/l CSF w.)
Moo	23	♀	W	35.5	7.41	24.1	46.8	7.33	23.1
Car	6	♀	N	39.4	7.39	25.5	49.8	7.32	24.4
Dav	22	♂	N	27.5	7.42	19.0	44.0	7.32	21.3
Mul	22	♀	W	28.0	7.45	20.9	51.6	7.30	24.0
Sto	14	♀	W	36.2	7.34	21.0	43.9	7.34	22.5
Fue	14	♂	W	40.5	7.39	26.4	50.6	7.31	24.0
Stu	10	♀	N	27.0	7.40	18.0	40.3	7.36	21.6
Sch	18	♀	W	39.0	7.38	25.9	42.8	7.38	24.2
War	20	♂	N	28.9	7.44	19.7	55.8	7.29	25.0
Gio	7	♂	W	29.3	7.40	18.2	39.6	7.37	21.6
Bro	69	♀	N	32.8	7.34	19.0	44.5	7.33	22.1
		Mean values		33.1	7.396	21.6	46.3	7.332	23.1
		S.D. (obs.)		±4.8	±.036	±3.2	±5.1	±.028	±1.3
		S.D. (mean)		±1.5	±.011	±1.0	±1.5	±.009	± .4
		Range		13.5	.11	8.4	16.2	.09	3.7

of normal values for blood acid-base status in such infants, serial examination of arterial blood was carried out for 3 consecutive days in each infant prior to study of CSF in order to assure that a steady-state of systemic acid-base equilibrium was present. Any infant whose values for whole blood buffer base for the first two days differed by more than 3 mEq/l from each other was excluded. Of the remaining infants a 2-way analysis of variance with respect to all acid-base variables of blood (*i.e.*, blood pH, plasma pCO_2, whole blood buffer base, whole blood base excess, and plasma bicarbonate) for each infant revealed no significant day to day differences over the 3-day period. All infants had normal body temperatures at the time of the study. Clinical data on the 13 children are shown in Table II. Many of these had some disorder of the neuromuscular system but none had hydrocephalus or active inflammatory disease of the brain or meninges. No patient had clinical or chemical evidence of pulmonary, renal or other disease known to affect acid-base equilibrium and all had values for blood acid-base variables within the normal range for their age(3,4).

Chemical methods. Arterial or arterialized capillary blood was analyzed for acid-base composition by methods previously reported (3,4,5). Plasma bicarbonate was calculated from the Henderson-Hasselbalch equation using a value of 0.0301 for S (the CO_2 solubility factor) and the appropriate value for pK'(6). Plasma pH was assumed to exceed blood pH by 0.010 pH units(6). Plasma water was assumed to be 93% of plasma.

Cerebrospinal fluid was obtained from an atraumatic lumbar puncture. CSF pH was measured in the Radiometer AME-1 instrument which was standardized with buffers made up from phosphate salts obtained from the National Bureau of Standards. Initially, considerable difficulty was encountered in obtaining reproducible values for CSF pH probably in part because CSF contains almost no non-bicarbonate buffer thereby magnifying the effects of small losses of CO_2 which may be incurred during collection of CSF or in filling the electrode. To obviate this potential source of error, a new technique of filling the electrode was developed. This consisted of connecting the electrode directly to the hub of the lumbar puncture needle by inserting one end of a length of sterile polyethylene tubing directly into the bore of the needle and fastening the other end to the glass electrode. When properly placed, no air bubbles entered the column of fluid as it passed through the connector tubing and into the electrode. A further precautionary step, recommended by Siggaard-Andersen(4) for poorly buffered fluids, consisted of filling the electrode initially with a sample of CSF, allowing it to sit for 30 to 60 seconds and then discarding it. Following this "preincu-

bation" of the electrode, the pH of subsequent samples of the same CSF was rapidly measured and the results averaged. Using the above precautions the reproducibility of CSF pH readings compared favorably with those obtained for blood pH.

Following measurement of CSF pH, a sample of CSF (approximately 1.0 ml in infants and 2.0 ml in children) was collected anaerobically in a gas-tight syringe and analyzed in duplicate using either the Natelson microgasometer or the Van Slyke manometric apparatus. All samples of CSF were crystal clear to the eye and with the exceptions noted

TABLE II. Data in Children.

Patient	Age (yr)	Sex	Race	Diagnosis	Plasma			CSF			Cells/mm³
					pCO_2 (mm Hg)	pH	$[HCO_3^-]$ (mEq/l plasma w.)	pCO_2 (mm Hg)	pH	$[HCO_3^-]$ (mEq/l CSF w.)	
Deg	4	♂	W	Post encephalitis	34.5	7.45	26.0	48.8	7.31	23.3	Not done
War	6	♀	W	Convulsive disorder	39.6	7.38	24.9	44.2	7.33	22.3	Normal
Wal	3	♂	W	" "	40.0	7.41	26.9	49.1	7.30	22.8	"
Per	2	♂	W	Hypotonia	38.6	7.42	26.4	47.8	7.35	24.8	"
Gia	6	♂	W	Eosinophilic granuloma	39.7	7.40	26.4	51.2	7.28	22.4	"
Ros	3	♂	W	Convulsive disorder	38.8	7.41	26.4	53.6	7.28	23.6	"
Lud	1	♀	W	Amyotonia congenita	37.0	7.43	26.6	49.2	7.30	22.3	"
Pet	4	♀	W	Astrocytoma	30.5	7.43	21.5	50.2	7.32	24.2	77.
Cot	4	♂	W	Muscular dystrophy	39.0	7.40	25.6	52.6	7.27	22.8	Not done
Tre	12	♂	W	Pseudo-papilledema	34.0	7.42	23.8	53.6	7.26	22.6	Normal
Jon	7	♂	W	Primary tuberculosis	33.0	7.44	24.1	49.8	7.30	23.1	"
Kod	½	♂	W	Convulsive disorder	36.3	7.43	26.1	56.6	7.29	25.8	"
Gra	4	♀	W	Headaches	40.5	7.38	25.8	53.8	7.28	24.5	17.
				Mean values	37.0	7.415	25.4	50.8	7.298	23.4	
				S.D. (obs.)	±3.2	±.022	±1.5	±3.2	±.025	±1.1	
				S.D. (mean)	± .9	±.006	± .4	± .9	±.007	± .3	
				Range	10.0	.07	5.4	8.8	.09	3.5	

all had normal cell counts and normal values for protein. CSF acid-base data were calculated from the Henderson-Hasselbalch equation using values for pK′ and for S as recommended by Mitchell *et al*(7). CSF was assumed to be 99% water.

Results. The individual data on the acid-base status for concurrently collected arterial blood and CSF for each of the 11 infants are shown in Table I. At the time of study, all infants had blood pH values within the range of 7.34 to 7.45 with a mean value (with standard error of the mean) of 7.396 ± 0.011 for the entire group. These values of blood pH were associated with values for plasma pCO_2 and plasma bicarbonate concentration which are low by adult standards but which agree well with the values reported by Albert and Winters(3) in older infants.

With one exception, the CSF pH of each infant was less than the pH of the arterial plasma, the mean value for CSF pH for the entire group being 7.332 ± 0.009. CSF pCO_2 always exceeded arterial plasma pCO_2, the mean value for CSF pCO_2 being 46.3 ± 1.5 mm Hg compared to that of plasma of 33.1 ± 1.5 mm Hg. This finding suggests that grossly unsteady states with respect to pCO_2 were not present, since it has been shown by others(8) that CSF pCO_2 corresponds closely to the pCO_2 of internal jugular venous plasma. In a steady-state, therefore, the latter must always be higher than the arterial plasma pCO_2 since the brain is adding CO_2 to the blood which enters it. CSF bicarbonate concentration showed no consistent directional deviation from that of arterial plasma water with the former exceeding the latter in 7 of 11 patients. The mean CSF bicarbonate concentration was 23.1 ± 0.4 mEq/l while that of arterial plasma water was 21.6 ± 1.0 mEq/l. Since the design of the study (see *Methods*) was such as to exclude patients experiencing major changes in the metabolic component of acid-base equilibrium of blood in the foreperiod, these values for CSF bicarbonate likely represent a reasonable approximation of the steady-state.

Table II shows individual blood and CSF acid-base data on the 13 children who were studied. The acid-base status of the blood of each of these subjects was within the normal range expected for age(3,4). As in the case of the infants, CSF pH was consistently lower than that of arterial plasma pH, and CSF pCO_2 was always greater than the pCO_2 in arterial plasma, the mean values being 7.298 ± 0.007 for CSF compared to 7.415 ± 0.006 for arterial pH and 50.8 ± 0.9 mm Hg for CSF pCO_2 compared to 37.0 ± 0.9 mm Hg for plasma pCO_2. In contrast to the infants, however, only one of the 13 values for CSF bicarbonate concentration exceeded the respective bicarbonate concentration in arterial plasma water, the mean value for CSF bicarbonate being 23.4 ± 0.3 mEq/l while that for arterial plasma water was 25.4 ± 0.4 mEq/l.

Table III summarizes data on the present study along with selected data§ from the literature representing normal values for 2 sizable series of adult subjects studied by acceptable methods. No statistically significant differences were found between any 2 of these groups with respect to blood pH. Both plasma pCO_2 and concentration of bicarbonate in arterial plasma water of the infants, however, were reduced to a significant extent when compared to either children or adults. CSF pCO_2 of the infants was also significantly lower than that of either children or adults but CSF bicarbonate concentration was not significantly different in the infants. Therefore, the CSF pH of the infants was higher to a slight but significant extent. The physiological significance of this difference is not clear, particularly since the difference is small

§ A complete summary of the reported data for normal acid-base composition of adult CSF will be found in the recent publication of Mitchell *et al*(1). In some of the previous reports mean CSF pCO_2 was less than that found by Mitchell *et al*(1) or Bradley and Semple(8) (shown in Table III). It is not unlikely that the lower values represent methodological errors of the type alluded to in *Methods*. Such a view is given further weight by the findings of Kety and Schmidt (9) that the pCO_2 of the internal jugular venous bulb plasma of normal adults (which is generally believed to be close to if not identical with the pCO_2 of CSF) averages 50 to 54 mm Hg. It is therefore difficult to accept values for CSF pCO_2 of normal adults which are significantly less than these values.

TABLE III. Summary of Normal CSF Data.

Age group	No. of subjects		pH Plasma	pH CSF	pCO_2 (mm Hg) Plasma	pCO_2 (mm Hg) CSF	$[HCO_3^-]$ (mEq/l water) Plasma	$[HCO_3^-]$ (mEq/l water) CSF
Infants	11	Mean values	7.396	7.332	33.1	46.3	21.6	23.1
		S.D. (obs.)	±.036	±.028	±4.8	±5.1	±3.2	±1.3
		S.D. (mean)	±.011	±.009	±1.5	±1.0	±1.0	± .4
Children	13	Mean values	7.415	7.298	37.0	50.8	25.4	23.4
		S.D. (obs.)	±.022	±.025	±3.2	±3.2	±1.5	±1.1
		S.D. (mean)	±.006	±.007	± .9	± .9	± .4	± .3
Adults:								
Mitchell *et al*(1)	12	Mean values	7.409	7.326	39.5	50.2	25.1	24.8
		S.D. (obs.)	±.018	±.012	±1.2	±2.6	±1.2	±1.4
		S.D. (mean)	±.005	±.003	± .3	± .8	± .3	± .4
Bradley & Semple(8)	25	Mean values	7.397	7.309	41.1	50.4	25.3	23.1
		S.D. (obs.)	±.022	±.026	±3.6	±1.4	±1.8	±1.4
		S D. (mean)	±.004	±.005	± .7	± .3	± .4	± .3

and is of the same order as observed between the two groups of normal adults.

Discussion. The results of the present study provide provisional standards for normal CSF acid-base composition in infants and children. The significant feature of the infant CSF is the lower value for pCO_2 compared to children or adults. This is not surprising in view of the fact that the pCO_2 of the plasma of these infants is normally lower than that of older children or adults. Previous studies from this laboratory(3) have suggested that the lower than adult values for plasma pCO_2 and for plasma bicarbonate observed in healthy neonates are maintained at least up to the age of 24 months and are statistically different from the values observed in older subjects. Our studies would suggest that the CSF pCO_2 of patients up to 2 years of age may also be lower than normal, although this has not as yet been directly tested.

The mechanisms responsible for the low plasma and CSF pCO_2 in infants are not clear. The results of the present study do show that the stimulus responsible for the augmented ventilation (relative to the rate of CO_2 production), which must be responsible for the low pCO_2, cannot be ascribed to a lower pH in the lumbar CSF of the infant in relation to that of children or adults.

Summary. The acid-base composition of arterial blood and cerebrospinal fluid has been measured in 11 infants of low birth weight and in 13 children in order to establish normal values. The CSF data in children and the usually accepted normal data for adults were similar. The infants, however, showed a significant reduction in CSF pCO_2 when compared to either children or adults. This undoubtedly reflects the lower values for plasma pCO_2 which are typical of this age group.

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