

found only in the middle portions of the intestine of the water fleas, where absorption of the luminescent components of the products of digestion of the vibrios evidently takes place (Fig. 3).

The results of these experiments thus showed for the first time that water fleas feed on enteropathogenic cultures of NAG vibrios, digest them, and assimilate this food with the aid of their digestive enzymes. Water fleas, which inhabit reservoirs in enormous numbers, participate in their purification and play the role of biological scavengers. This property of these organisms can be utilized to good purpose by the antiepidemic and hygiene services for the prevention and control of intestinal infections.

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#### PERCENTAGE OF EAC-RFC AND T<sub>γ</sub> CELLS IN THE THYMUS AND SPLEEN OF HUMAN FETUSES

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The present stage of development of immunology is characterized by a penetrating and all-embracing study of receptors of immunocompetent cells (ICC), i.e., those unique structural formations by means of which the numerous cooperative interactions leading to different types of immunologic reactions take place [3, 4, 11, 14]. There have been many studies of the formation of receptors to ICC in the prenatal development of bone marrow [1, 6-8, 14, 15] and various animals [3, 9, 10]. Nevertheless, analysis of some of them [13] reveals considerable technical errors, which have led to the obtaining of incorrect data and, as a result of this, to an incorrect interpretation of the true state of affairs. The problem of formation of certain receptors such as, for example, the receptor for the Fc fragment of IgC on T lymphocytes of human fetuses is not mentioned at all in the literature, although the property of T suppressors has been ascribed to lymphocytes of this type [12].

In the investigation described below the percentages of EAC rosette-forming cells (EAC-RFC) and of T<sub>γ</sub> cells were studied in the thymus and spleens of human fetuses.

#### EXPERIMENTAL METHOD

Altogether 22 human fetuses aged 11-28 weeks, obtained as a result of spontaneous abortions from clinically healthy mothers not receiving any drugs were used. In two cases the abortions were criminal. Four suspensions from the thymus and spleen were obtained by the method described by Raitsina et al. [5]. The T lymphocytes were obtained from the spleen by the

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TABLE 1. Percentage of EAC-RFC and T<sub>γ</sub> Cells in Thymus and Spleen of Human Fetuses Obtained by Spontaneous Abortion

Age of fetuses, weeks	EAC-RFC		T <sub>γ</sub> cells	
	thymus	spleen	thymus	spleen
11-14	0.7±0.3 (0-1.5) n=5	5.5±4.5 (1-10) n=3	0.6±0.1 (0.5-1) n=3	1 — n=1
18-23	0.6±0.2 (0-1) n=6	17.0±4.3 (4-28) n=6	1.0±0.8 (0-5) n=7	3.9±1.1 (1-8) n=6
24-28	0.9±0.5 (0-4) n=7	20.7±3.8 (10-37) n=7	0.3±0.3 (0-1.5) n=5	5.6±0.6 (5-7.5) n=4

Legend. EAC-RFC) Lymphocytes with receptors for the 3rd fragment of complement; T<sub>γ</sub> cells) lymphocytes with receptors for the Fc fragment of IgG. n) Number of fetuses studied.

reaction of spontaneous rosette formation between human lymphocytes and sheep's red blood cells, followed by isolation of a pure E-RFC population on a Ficoll-Verografin density gradient [3]. The content of E-RFC purified by this method in spleen suspensions was 97-100%. The content of T lymphocytes in thymus suspensions, determined with the aid of specific anti-T-serum, kindly provided by N. A. Kraskina, was 98-100%. The EAC-RFC were determined by the spontaneous rosette formation test with human lymphocytes and sheep's red blood cells, loaded with antibodies and complement, the source of which was fresh serum from noninbred mice, diluted in the ratio of 1:10 [3]. To obtain T<sub>γ</sub> cells the method described by Gupta and Good [9] was used. Total Ig of the G class was obtained by ion-exchange chromatography on DEAE-cellulose from hypoimmune rabbit antiserum. The results of Ouchterlony's test and of immunoelectrophoresis in agar showed that the preparation thus obtained was pure total IgG free from contamination by Ig of other classes.

#### EXPERIMENTAL RESULTS

It will be clear from Table 1 that the content of EAC-RFC in the thymus of fetuses of different ages did not exceed on average 1%. These data agree with the results obtained previously by the writers on rat fetuses [3], and also by other workers using animals of other species [9]. They are diametrically opposite to the conclusions reached by some workers [13], who used sheep's red blood cells loaded with antierythrocytic antibodies and complement in order to determine EAC-RFC in the human fetal thymus. The possibility cannot be completely ruled out that T lymphocytes can easily bind directly with red blood cells, and that this may lie at the basis of the reaction of spontaneous rosette formation between human lymphocytes and sheep's red blood cells used for detecting human T lymphocytes. It is the spontaneous rosettes of fetal thymocytes with sheep's red blood cells that are not readily destroyed by heat, providing the necessary control for use when the red blood cells of animals of this species are used in the system of EAC rosette formation to detect C3-positive human lymphocytes. The content of EAC-RFC in spleen suspensions used as the control ranged on average from 5.5% at the 11th-14th weeks of development to 20.7% at the 24th-28th weeks, which correlates with the number of Ig<sup>+</sup>-cells in the spleen in the course of intrauterine development [6]. The very small number of C3-positive lymphocytes in the human fetal thymus is strikingly similar to the low content of Ig<sup>+</sup>-cells discovered by Chuich in this organ in the prenatal development of both man [6] and the rat [3].

It can be postulated on the basis of the facts described above that EAC-RFC in the human fetal thymus are represented not by T cells, but by B lymphocytes (Ig<sup>+</sup>-cells), of which a receptor for C3 is highly characteristic [2]. The proportion of T<sub>γ</sub> cells in the human fetal thymus does not exceed 1% at any of the times of development studied, the same as the proportion of T<sub>γ</sub> cells in the adult human thymus [8]. The content of the above-mentioned

cells in the spleen of the fetuses studied varied from 1 to 8%, much less than the 45% found in the adult human spleen [8]. In particular it must be emphasized that the proportion of these cells in the spleen of two fetuses at 21 and 25 weeks of development, obtained as a result of criminal abortions, was significantly higher, namely 18 and 31% respectively. It will be recalled that fetuses of this type reflect normal **intrauterine human development**. Olding and Oldstone [12], who studied the reaction of the peripheral blood lymphocytes of newborn infants and their mothers in mixed culture, showed that the T lymphocytes of the former inhibit blast transformation of the latter considerably; this may be one of the factors maintaining the normal course of pregnancy. These workers concluded that T lymphocytes of newborn infants responsible for this reaction are suppressors, for the killer activity of the cells was absent in cytotoxicity detection experiments. If the proposition is accepted that  $T_\gamma$  cells have a suppressor action on ICC [11], the low proportion of these cells compared with adult persons and fetuses obtained as a result of criminal abortion may evidently play a definite role in the pathogenesis of spontaneous abortion and miscarriages.

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