

40 μ , near anterior margin. Legs long and slender, tarsi 2-segmented. Fore wings (Fig. 3) ensiform, length 680 μ , width 35 μ at about middle. Costa with 25 setae. Upper vein with 14 setae in 2 groups and 2 apical setae. Lower vein with 12 setae in about equal space. Anal lobe with 5 setae at anterior margin and 1 seta near base at middle.

Abdominal tergites without sculpture: I-IX with a prominent transverse line near anterior margin; I-VIII with median pair of small setae, ahead of median discal pore; IX (Fig. 4) with 4 pairs of setae, i, 26 μ , ii; 15 μ , iii; 13 μ ; iv. 47 μ ; X with 1 pair of major setae, 85 μ . Abdominal sternites weekly sculptured, without accessory setae.

Female: Unknown.

Holotype: male, from *Pinus massoniana* L., Taoyuan, Kaohsiung Hsien, Taiwan; VII 6, 1975, L. S. Chen. The type-specimen deposited in the collection of Plant Quarantine Division, Bureau of Commodity Inspection and Quarantine, Ministry of Economic Affairs, Taipei, Taiwan, Republic of China.

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REFERENCES

1. Hood, J. D. (1915). An interesting case of antennal antigeny in Thysanoptera. *Proc. Ent. Soc. Washington* 17(3): 128-132.
2. Hood, J. D. (1956). A new genus and species of Thripidae (Thysanoptera) from Trinidad. *Proc. R. Ent. Soc. London* (B), 25: 64-66.
3. Stannard, L. J. (1968). The Thrips, or Thysanoptera, of Illinois. III. *Hist. Surv. Bull.* 29(4): 209-552.

臺灣薊馬科之一新屬及新種

陳 連 勝

毛角曾氏薊馬 (*Tsengothrips plumosa*), 在高雄縣桃源之馬尾松上採得, 爲屬於薊馬科之一新屬新種。

本屬 (*Tsengothrips*) 與 *Plesiothrips* 和 *Plesioopsotrips* 極相似, 然以頭部呈梯形, 觸角第三節短, 中單眼位於二複眼之間, 三個單眼大及二個後單眼略呈三角形, 可和後兩屬區別。

Blood and Cerebrospinal Fluid Findings in Eosinophilic Meningitis and Antibody to *Angiostrongylus cantonensis*

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An outbreak of an unusual form of meningitis, characterized by a pleocytosis (in particular, eosinophilic leucocytes), was recognized in Taiwan. The disease have been referred to as eosinophilic meningitis or meningoencephalitis. Recently, it has been reported that this disease can also be caused from the infection by the 3rd stage larvae of *Angiostrongylus cantonensis*. In man who serves as an accidental host of this parasite, the larvae may migrate through the central nervous system. The clinical manifestations as well as immune response of the body may occur during the larval migration. In the present investigation, the levels of immunoglobulin, antibody to *A. cantonensis* antigen and the number of leucocytes in serum and cerebrospinal fluid (CSF) from the patients of eosinophilic meningitis are investigated in order to seek the evidence for the presence of a specific immune response to the pathogenic stimulation.

MATERIALS AND METHODS

Five patients of eosinophilic meningitis all had past experience of eating raw *Achatina fulica* within a month prior to the onset of the disease. From each patient 3 ml of serum and cerebrospinal fluid (CSF) were obtained and stored in a deep freezer

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(-70°C) until used. Meanwhile, the thin smears of peripheral blood and of the sediment of CSF were made, and the white blood cells (WBC) were classified and counted after staining with Giemsa's solution. For comparison, blood and CSFs were also collected from twenty healthy persons.

Immunoglobulin and Total Protein Determination: The quantitation of IgG, IgE, IgA, IgD and IgM in sera and CSFs were done by the radial immunodiffusion method of Mancini⁽¹⁾ by using commercially available immunoplates (Hyland, Los Angeles, California). All measurements were performed in duplicate. Total protein concentration in CSFs was determined by the Folin method⁽²⁾.

Preparation of Antigen and Antibody Detection: The preparation of antigen employed for the indirect hemagglutination (IHA) test was summarized schematically in Fig. 1. From the whole worm extract of *A. cantonensis*, several cross reacting components were removed by DEAE-cellulose column chromatography with stepwise elution with different concentration of phosphate buffered saline (PBS)⁽³⁾. Subsequently, components common to rat serum were also absorbed by passing through a column of the cyanogen bromide-activated sepharose 4B (Pharmacia Fine Chemicals) coupled with rabbit immunoglobulin against normal rat serum as described by Sato *et al.*⁽⁴⁾. With purified antigen, the antibodies

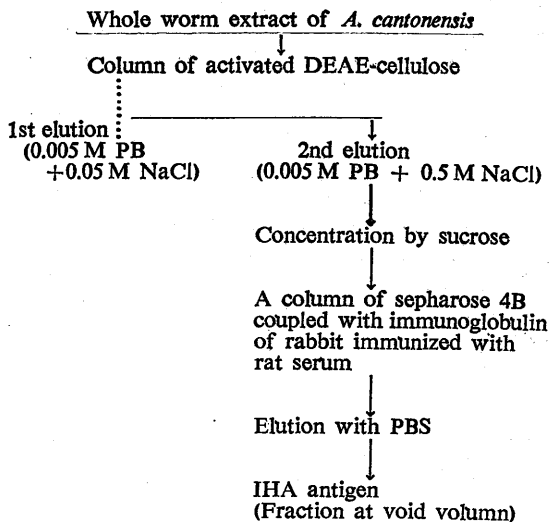


Fig. 1. Procedure of the preparation of *Angiostrongylus cantonensis* antigen for the indirect hemagglutination (IHA) test

in four sera of the patients were tested by IHA test of Jacobs and Lunde⁽⁵⁾ with a minor modification as reported in the previous report⁽⁶⁾. In addition, the double immunodiffusion was also accomplished in all of the sera with crude worm extract⁽⁷⁾.

RESULTS

The number of WBC and the levels of various components of immunoglobulins in the peripheral bloods and CSFs from sick and normal persons were compared and the results were presented in Table 1 and 2. From Table 1, it was found that the amount of IgM and IgE were higher in the patients sera than those in the normal sera. However, no changes of the amount of sera IgA, IgD and IgG on the outbreak of eosinophilic meningitis were found. As far as the amount of various components of immunoglobulins in CSF was concerned, the data (Table 2) revealed that the amount of IgG, IgA and IgM increased in the patients of eosinophilic meningitis.

In addition to the change of the amount of sera and CSF immunoglobulin in the patients, the number of WBC also increased both in blood and CSF. Furthermore, the total protein concentration in CSF also increased following pathogenic infection.

The titers of antibody against the purified *A. cantonensis* antigen were 1:128 in two patients and 1:256 in the other two sera. By using double immunodiffusion, all the sera of five patients gave a visible precipitin reaction to the crude worm antigen.

DISCUSSION

Tungkanak *et al.* reported that the protein pattern of the CSF in the patients with eosinophilic meningitis was significantly different from that of CSF of the control subjects. Similarly, the present investigation also showed that the CSFs of the patients revealed significantly higher levels of IgG, IgA, IgM and total protein concentration in CSFs. Usually, it is difficult to find any immunoglobulin from normal CSF by using immunoplate. However, with the same method, IgG, IgA and IgM levels of immunoglobulin are easily detected from the CSFs in the patients with eosinophilic meningitis. From the present data, the increases in the CSF IgG and IgA levels were proportionally higher than the increases of the corresponding serum immunoglobulins. It may be an indirect piece of evidence for local immunoglobulin synthesis in the central

Table 1. Number of leucocyte, immunoglobulin levels and antibody to *Angiostrongylus cantonensis* in blood from eosinophilic meningitis patients

Case No.	WBC ($\times 11/9$) mm ³	Leucocyte (%)			Immunoglobulin (Mg/100 ml)					IgM	Double Immunodiffusion (with crude antigen)	IHA titer (with purified antigen)
		Mono.	Eosino.	Lymph.	IgG	IgA	IgD	IgE (IU ₁ /100ml)				
1	13,950	2	15	21	1100	200	2.6	1470	195	+	1:128	
2	15,700	1	20	26	1400	150	17.0	1538	285	+	1:128	
3	10,500	5	8	16	1300	310	7.0	1580	580	+	1:256	
4	18,700	2	30	31	1300	330	5.8	NM**	700	+	1:256	
5	17,800	6	8	42	740	200	4.9	1400	285	+	NM	
Mean \pm S.D.					1286 \pm 348.9	238 \pm 69.9	7.5 \pm 4.9	1508 \pm 77.7	409 \pm 195.2			
Control					1319 \pm 268.2	268 \pm 86.3	7.4 \pm 4.2	1280 \pm 322.4	122.6 \pm 38.8			
t					0.20	0.82	0.04	2.85	3.26			
(P)					(>0.5)	(>0.4)	(>0.5)	(<0.01)	(<0.01)			

*: Positive reaction

** : Not measured

† : International unit

t: t distribution to show significance of difference between two successive means.

t_{0.01} = 2.807, significant level P: probability

Table 2. Number of leucocytes and immunoglobulin levels in CSF from eosinophilic meningitis patients

Case No.	WBC ($\times 11/9$)	Leucocyte (%)			Total protein	Immunoglobulin (mg/100 ml)				
		Mono.	Eosino.	Lymph.		IgG	IgA	IgD	IgE	IgM
1	1396	2	75	20	52	—*	19	—	—	25
2	1658	1	84	15	60	64	20	—	—	18.5
3	1520	2	40	20	85	20	10	—	—	19
4	1546	0	95	2	65	25	20	—	—	20
5	458	4	28	66	45	52	20	—	—	18.5
Mean \pm S.D.					61.4 \pm 11.9	40.3 \pm 10.3	17.8 \pm 3.9	—	—	20.5 \pm 2.5
Control					36.5 \pm 8.4	4.0 \pm 0.8	0.7 \pm 0.2	—	—	—
t					4.41	7.87	11.86	—	—	16.4
(P)					(<0.01)	(<0.01)	(<0.01)			(<0.01)

*: Not detectable

** : Not measured

t: t distribution to show significance of difference between two successive means.

t_{0.01} = 2.807, significant level P: probability

nervous system (CNS) in response to the pathogen of the eosinophilic meningitis. Similarly, same immune response of CNS to larvae migration has been quoted by Tungkanak *et al*⁽⁹⁾. Interesting enough, a very highly significant increase of IgE and IgM was found in the sera of the patients. It is known that IgE (or reagin) is correlated with hypersensitivity or allergy caused by the pathogens. Increased concentration of IgM class antibody has been documented in patients infected with protozoa, leptospirae, rickettsiae and helminths^(9,10,11). Under such circumstances, the high level of IgM of the patient may indicate that the disease was caused by the parasitic infection. As far as the increasing of IgE is concerned, it may be referred to that the symptom of the eosinophilic meningitis may be caused, in part, by immediate hypersensitivity induced during the course of the sickness.

From the present investigation, the significantly higher IgG, IgA, IgM levels and total protein concentration in CSF as well as IgE and IgM in the serum of eosinophilic meningitis may be taken as an sufficient piece of evidence for indicating the specific immune response of eosinophilic meningitis.

The present study showed that the increase of eosinophils was more significant in CSF than those in blood. Therefore, the increase of CSF eosinophils seems to be a better indication of eosinophilic meningitis. Considering all the immune responses of human subjects after infection of *A. cantonensis*, by using fractionation and immunoadsorbent techniques, a specific *A. cantonensis* antigen was obtained and utilized for the detection of antibody in serum. Four tested sera from eosinophilic meningitis patients showed high IHA titers ($\geq 1:128$). With double immunodiffusion, the sera reacted with the crude *A. cantonensis* extracts. The data also showed that the higher IHA titer may be related to the IgM immunoglobulin of sera of the patients (Table 1). With the history of eating raw *Achatina fulica* and higher IHA titers as well as positive reaction in precipitation test, it is possible that the cases of eosinophilic meningitis studied in this report may be caused by *A. cantonensis*.

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LITERATURE CITED

1. Mancini, G., A. C. Carbonara and J. F. Heremans, 1965. Immunochemical quantitation of antigens by single radial immunodiffusion, *Immunochemistry*, 2: 513-526.
2. Kabat, E. A. and M. M. Mayer, 1961. *Experimental Immunochemistry*, 2nd ed., Charles C. Thomas, Springfield, III, p. 556-558.
3. Cheng, L. Y., J. K. Chiu and T. Suzuki, 1974. Studies on immunodiagnosis 2. Preliminary experiment for preparing specific antigen from *Angiostrongylus cantonensis*. *J. Parasitol.*, 23: 35-41.
4. Sato, Y., T. Suzuki, T. Yamashita, H. Sekikawa, M. Otsuru, S. N. Chen, S. Y. Lee and K. H. Liu, 1974. Studies on immunodiagnosis of angiostrongyliasis. 4. Purification of antigen by using immunoadsorbent. *Jap. J. Parasitol.*, 23: 42-45.
5. Jacobs, L. and M. N. Lunde, 1957. A hemagglutination test for toxoplasmosis. *J. Parasitol.*, 43: 308-314.
6. Chen, S. N. and T. Suzuki, 1974. Fluorescent antibody and indirect hemagglutination test for *Angiostrongylus cantonensis* infection in rats and rabbits. *J. Formosan Med. Associa.*, 73: 393-400.
7. Chen, S. N. 1974. Studies on immunodiagnosis of angiostrongyliasis 5. Double immunodiffusion and counterelectrophoresis for detection of antibody in infected rat and human sera. *Chin. J. Microbiol.*, 7: 114-118.
8. Tungkanak, R., S. Sirisinha and S. Punyagupta, 1972. Serum and cerebrospinal fluid in eosinophilic meningoencephalitis: Immunoglobulins and antibody to *Angiostrongylus cantonensis*. *Am. J. Trop. Med. Hyg.*, 19: 950-958.
9. Fahey, J. L. 1965. Antibodies and immunoglobulins. Normal development and changes in disease. *JAMA*, 194: 141-144.
10. Konovalova, L. M. and O. G. Poletova, 1974. Determination of immunoglobulin types in the serum of patients with cerebral cysticerciasis. *Meditsinskaya Parazitologiya Parazitarnye Bol-ezni*, 43: 285-289.
11. Bassily, S., Z. Farid, M. I. Saied, F. Iskander, Y. Mahran and M. I. Moussa, 1973. Serum immunoglobulin levels in different stages of *S. mansoni* infection and the effect of therapy on these levels. *J. Egypt. Med. Associa.* 56: 433-439.

嗜酸性腦炎病患血液及腦脊椎液中
白血球、免疫球蛋白之變化及
對廣東住血線蟲之抗體

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本研究乃從五位患嗜酸性腦炎之病人中取得血清及腦脊椎液測定其蛋白質成分之變化以及對廣東住血線蟲抗原之血清反應，據此以了解臺灣的嗜酸性腦炎患者之免疫反

應。結果顯示在病人腦脊椎液內之 IgG, IgA, IgM 和全蛋白濃度及血液中的 IgE 及 IgM 有顯著的增加。以雙相擴散反應測定抗體之存在時，則病人皆有沉澱反應。應用間接血球凝集法時可測出病患之抗體效價達到 256 倍。於腦脊椎液中的偏高免疫球蛋白顯示出中樞神經系統對病原的刺激發生合成抗體之反應。實驗結果亦顯示出高的抗體效價可能與血清 IgM 免疫球蛋白有關。除此而外，所有病人的血清及腦脊椎液中白血球總數及嗜酸性白血球皆顯著的升高。