

# Study on Histomorphology and Quantitative Variations of 'COMMA HAIRS' of Esperto in Samples of Papers

J. Bhattacharyya, Miss Gita Guha and B. Bhattacharyya  
Central Forensic Science Laboratory, Calcutta

*10,000 (ten thousand) 'comma hairs' (trichomes) of esperto are microscopically detected in samples of paper and examined histomorphologically. They are arbitrarily classified by the authors into ten types pointing out their distinguishing features. These types are later traced with camera lucida and photographed. They are, however, subjected to quantitative analysis such as percentage occurrence, mean, standard error of the mean, standard deviation, co-efficient of variation, student's 't' test and Carl Pearson's co-efficient of correlation(r). The variations in 'comma hairs' or trichomes are found to be of great diagnostic value. The result hits out a possibility in detection of crime.*

## Introduction

Esperto (*Stipa tenacissima*) is an important pulp used in high grade printing papers.<sup>1</sup> Paper industry utilises the blades (leaves) of esperto grass for manufacture of pulp. When examined under the microscope, esperto is found to be composed of (i) sharply pointed fibres, (ii) serrated epidermal cells, (iii) vascular tissue and (iv) 'comma hairs' or trichomes. These trichomes are found to occur abundantly in esperto. The process of pulping does not destroy these trichomes and their histomorphological features are retained very much intact due to the reason that the walls of trichomes are of cellulose impregnated with silica or calcium carbonate having a protective covering a cuticle.<sup>2</sup> The trichomes vary in shape, size and distribution.<sup>3</sup> Presence of esperto in a sample of paper affords useful clues in the detection of crime in that expert opinion is often needed for the identification of paper, antique collection of documents and so on. In the present work the trichomes are analysed on all counts histomorphologically and

statistically to bring out their salient features.

## Materials

Available paper samples :

1. Printing M.F. Toned, 20"x30", 500 sheets to ream, 30 lb, Umbrian, Messrs. Spicers Ltd., New Bridge Street, E.C. 4.
2. Art Two sided tinted, 23"x36", 500 sheets to Ream, 70 lb, vellum, allied paper merchants, Mansell Street, E.1.
3. Offset Cartridge, White, 20"x30", 500 sheets to Ream, 48 lb, W. S. L., Messrs. Lepard and Smiths Ltd., Earlhon Street, W.C. 2.
4. Immitation Art toned 20"x30", 500 sheets to Ream, 48 lb. W.S.L., Messrs. Lepard and Smiths Ltd., Earlhon Street, W.C. 2.

## Experimental Methods :

Papers were torn into pieces, kept in a test tube, boiled in 1% sodium hydroxide solution (aqueous) for 10-15 minutes,

washed in distilled water neutralized in 0.05 N HCl and washed again and again in distilled water. The whole material was converted into pulp. A small quantity of the pulp thus made was rolled between fingers to form pellets, kept in a test tube and shaken vigorously with little quantity of distilled water till the whole material is defibred. Distilled water was further added later on to obtain a fibre concentration of 0.05% to 0.1%. One or two drops of fibre suspension was taken on a glass slide, covered with cover glass and examined under the microscope. Only 'comma hairs' or trichomes were studied for 10,000 counts. They were classified into ten types by the authors according to their histomorphological variations. The distinguishing features were traced with camera lucida and photographed.

## Statistical Methods :

After the typing of the trichomes, percentage occurrence of each type within a sample of paper was studied. A study on the incidence of all the ten types of trichomes was conduc-

ted and various statistical constants such as mean, standard error of the mean, standard deviation and co-efficient of variation were worked out on the basis of suitable statistical models. Student's 't' test of statistical significance between the percentage occurrence of the different types of trichomes was performed. In addition, Karl Pearson's co-efficient of correlation (r) between various types of trichomes occurring in samples of esperto was studied.

### Histomorphology

It can be seen from the plate 1 (fig. 1 to fig. 33) that there are ten types of trichomes occurring in esperto. They are as follows: TYPE : I. Unicellular, thick walled, strong cuticular warts, tip blunt and base flask-shaped (fig. 1-3).

TYPE : II. Non-glandular, unicellular, simple, varying in length, diameter and thickness, stiff, conical, some short, bent, pointed or tip mostly pointed, base flat or irregular, or angular or broken (fig. 4-12).

TYPE : III. Short basal cell and a long terminal cell. Basal cell with silicified walls having the appearance of dark curves or translucent dots; tips wollen, sub-terminal region concave (fig. 13-15).

TYPE : IV. Small, pear-shaped, unicellular, hooked at the apex, acuminate, moderately thick-walled; thick protoplasmic inclusions in the basal region (fig. 16-18).

TYPE : V. Tip typically beaked, conical, base swollen and flat. invaginations at the tip region, prominent (fig. 19-21).

TYPE : VI. Tip hooked, projected, base swollen, bottle-shaped with protoplasmic inclusions; thick cuticular appearance in the terminal portion (fig. 22 & 23).

TYPE : VII. Stiff, tip pointed, sub-terminal region bent and widened, base flat, round, mid region concave; thick striation marks parallel (fig. 24 & 25)

TYPE : VIII. Unlike trichomatous appearance, unicellular with external blunt protruberance, small, tip blunt or notched, base swollen and flat; base with vacuolated protoplasmic mass (fig. 26-28).

TYPE : IX. Bent, tip blunt, bottle-tip appearance, base swollen containing silica particles (fig. 29-30).

TYPE : X. Deformed, unicellular, apices thickwalled, lateral walls wavy, tip protruded and blunt, sub-terminal region highly constricted; base crenulated, uncommon or even absent (fig. 31-33).

### Quantitative Variations :

The results of various statistical analyses are indicated in figures 2 and 3 and tables 1 to 4.

TABLE 1.  
Percentage incidence of different types of trichomes in esperto of paper :

| Types of trichomes | n    | % $\pm$ S.E.      |
|--------------------|------|-------------------|
| I                  | 1000 | 3.06 $\pm$ .2966  |
| II                 | 1000 | 56.07 $\pm$ .3242 |
| III                | 1000 | 7.99 $\pm$ .7351  |
| IV                 | 1000 | 5.97 $\pm$ .5613  |
| V                  | 1000 | 5.81 $\pm$ .5473  |
| VI                 | 1000 | 9.93 $\pm$ .8943  |
| VII                | 1000 | 0.07 $\pm$ .2027  |
| VIII               | 1000 | 5.07 $\pm$ .4814  |
| IX                 | 1000 | 3.04 $\pm$ .2947  |
| X                  | 1000 | 0.99 $\pm$ .0980  |

The following deductions could be advanced on the basis of study on trichomes :

1. Percentage occurrence of each type of trichome was distinctly different from the others as no two types were found to occur identically.

2. Of all the types of trichomes, type II showed the highest incidence ( $56.07 \pm .3242$  percent) and the type X showed the lowest incidence ( $0.99 \pm .098$  percent).

3. The result of 't' test between mean percentage of different types of trichomes revealed that the type I failed to establish significant difference with either of the types VII and IX; so also was observed in type III when studied with each of the types IV, V and VI, type IV with each of the types V and VIII, type V with type VIII and type VII with each of the types IX and X. All other values of 't' were found to reach the level of statistical significance.

4. The range of occurrence of trichomes was found highest with regard to type II (5357—5846) and least in type IX (255—342).

5. Studies on correlation showed higher values of 'r' between types V and VI (.9358), types V and VIII ('r'=.8728), types II and VIII ('r'=.8728), types II and VIII ('r'=.6060) and types III and VI ('r'=.4841). Hence in every sample of esperto all the above types of trichomes are likely to show their abundance. With regard to the remaining types (I, IV, VII, IX and X) of trichomes, although they are of very high diagnostic value, yet may or may not occur in every sample.

TABLE 2

Student's 't' test of significance between the Percentage occurrence of different types of trichomes :

|      | II                  | III               | IV               | V                 | VI                | VII              | VIII             | IX                | X                 |
|------|---------------------|-------------------|------------------|-------------------|-------------------|------------------|------------------|-------------------|-------------------|
| I    | (53.01)<br>67.2801* | (4.93)<br>4.85*   | (2.91)<br>3.14*  | (2.75)<br>2.99*   | (6.87)<br>6.30*   | (0.99)<br>1.40   | (2.01)<br>2.28*  | (0.02)<br>0.03    | (2.07)<br>3.30*   |
| II   | —                   | (48.08)<br>46.72* | (50.1)<br>53.24* | (50.26)<br>53.84* | (46.14)<br>41.80* | (54.0)<br>74.39* | (51.0)<br>56.82* | (53.03)<br>67.41* | (55.08)<br>84.76* |
| III  | —                   | —                 | (2.02)<br>1.77   | (2.18)<br>1.93    | (1.94)<br>1.52    | (5.92)<br>6.11*  | (2.92)<br>2.09*  | (4.95)<br>4.88*   | (7.0)<br>7.67*    |
| IV   | —                   | —                 | —                | (0.16)<br>0.95    | (3.96)<br>3.28*   | (3.90)<br>4.46*  | (0.90)<br>0.97   | (2.93)<br>3.17*   | (4.98)<br>6.13*   |
| V    | —                   | —                 | —                | —                 | (4.12)<br>3.43*   | (3.74)<br>4.32*  | (0.74)<br>0.73   | (2.77)<br>3.03*   | (4.82)<br>6.03*   |
| VI   | —                   | —                 | —                | —                 | —                 | (7.86)<br>7.50*  | (4.86)<br>4.14*  | (6.89)<br>6.32*   | (8.94)<br>8.98*   |
| VII  | —                   | —                 | —                | —                 | —                 | —                | (3.0)<br>3.63*   | (0.97)<br>1.38    | (1.08)<br>1.97    |
| VIII | —                   | —                 | —                | —                 | —                 | —                | —                | (2.03)<br>2.30*   | (4.08)<br>5.36*   |
| IX   | —                   | —                 | —                | —                 | —                 | —                | —                | —                 | (2.05)<br>3.27*   |

Figures in ( ) indicate the difference between mean.

\* Significant.

TABLE 3

Statistical constants for the occurrence of different types of trichomes in samples of esperto of paper :

| Types of trichomes : | Range     | Mean $\pm$ S.E.      | S.D.   | C.V.  |
|----------------------|-----------|----------------------|--------|-------|
| I                    | 201— 394  | 302.0 $\pm$ 3.1123   | 53.91  | 17.85 |
| II                   | 5357—5846 | 5600.93 $\pm$ 1.8034 | 134.96 | 24.10 |
| III                  | 609— 987  | 798.5 $\pm$ 3.3695   | 95.31  | 11.94 |
| IV                   | 413— 788  | 595.92 $\pm$ 4.2403  | 103.87 | 17.43 |
| V                    | 403— 744  | 579.09 $\pm$ 3.4358  | 84.16  | 14.53 |
| VI                   | 811—1179  | 988.05 $\pm$ 2.8618  | 90.50  | 9.16  |
| VII                  | 151— 247  | 00.0 $\pm$ 1.7674    | 24.99  | 12.50 |
| VIII                 | 307— 686  | 501.5 $\pm$ 4.5504   | 101.75 | 20.29 |
| IX                   | 255— 342  | 301.16 $\pm$ 1.4428  | 24.99  | 8.30  |
| X                    | 4— 148    | 99.0 $\pm$ 3.5675    | 35.68  | 36.04 |

## Acknowledgement :

Thanks are due to Dr. M. Jauhari, Director, Central Forensic Science Laboratory, Calcutta for constant encouragement, guidance and facilities. Thanks are also due to Shri B. Manna, Scientific Assistant for photographic work and to Shri Ramnath Singh, Laboratory Attendant, for technical assistance.

## References

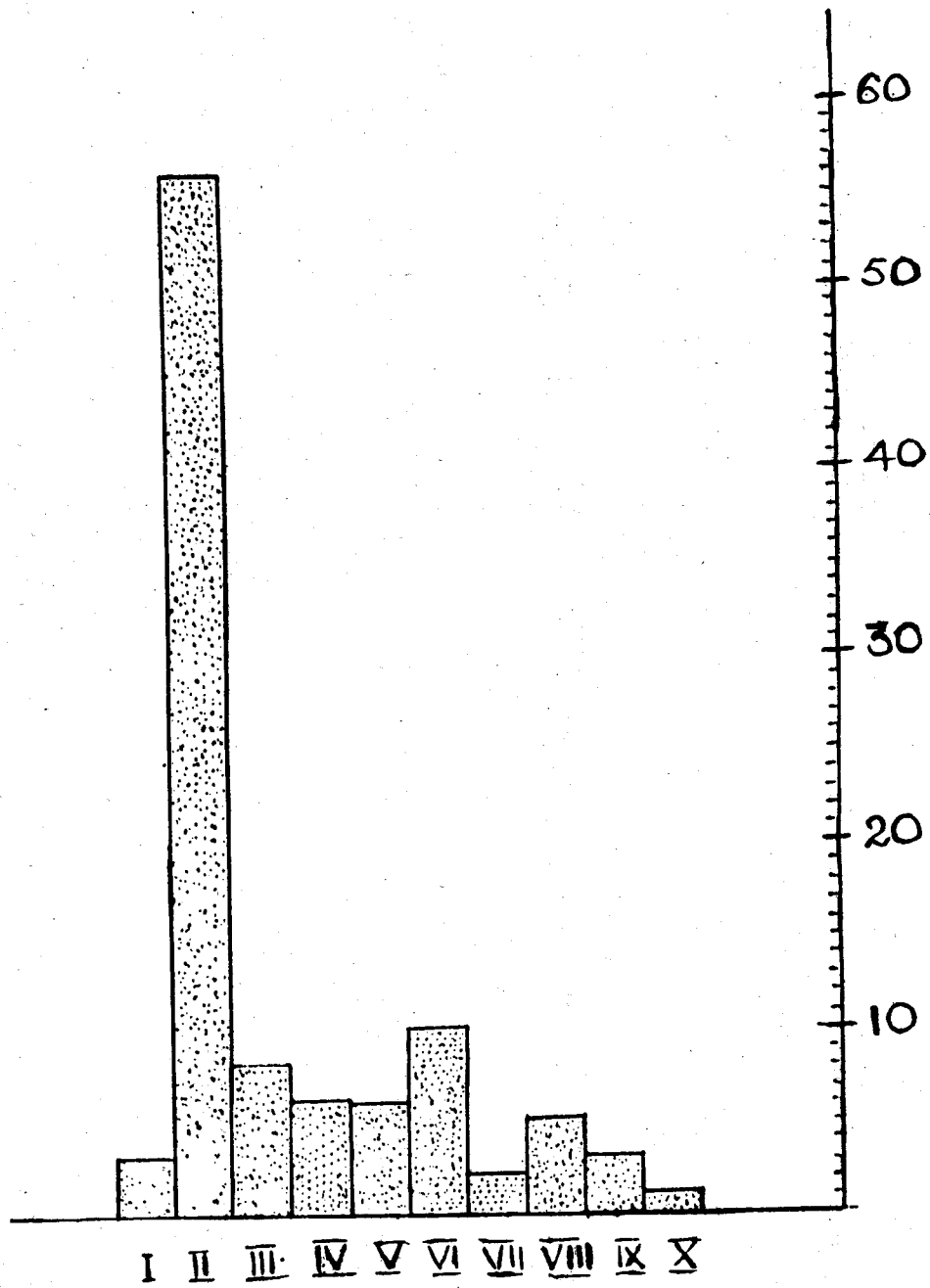
1. Casey, J. P. (1952) Pulp and Paper Chemical Technology, Vol. I, Interscience.
2. Beyrich, H. (1943) Uber die Membranver Kieselung einiger Pflanzenhaare. Flora 36 : 313-324.
3. Esau, K. (1953) Plant Anatomy. John Wiley & Sons, Inc. N.Y. : p. 153.

TABLE 4

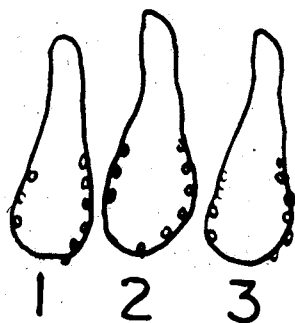
Studies on Correlation ( $r$ ) between various types of trichomes on the basis of their occurrence in various samples of esperto of paper :

| Between types : | X    | Y    | X <sup>2</sup> | Y <sup>2</sup> | XY    | $r \pm S.E.$ |                    |
|-----------------|------|------|----------------|----------------|-------|--------------|--------------------|
| I & II          | 60   | 1120 | 198            | 64388          | 3381  | +            | .12128 $\pm$ .518* |
| I & III         | 60   | 160  | 198            | 1406           | 466   | -            | .2940 $\pm$ 1.31*  |
| I & IV          | 60   | 120  | 198            | 776            | 363   | +            | .0945 $\pm$ .403   |
| I & V           | 60   | 120  | 198            | 778            | 358   | -            | .0619 $\pm$ .263   |
| I & VI          | 60   | 200  | 198            | 2126           | 602   | +            | .0420 $\pm$ .178   |
| I & VII         | 60   | 40   | 198            | 102            | 122   | +            | .1005 $\pm$ 1.01*  |
| I & VIII        | 60   | 100  | 198            | 550            | 307   | +            | .2333 $\pm$ 1.02*  |
| I & IX          | 60   | 60   | 199            | 208            | 177   | -            | .1336 $\pm$ .572*  |
| I & X           | 60   | 20   | 198            | 22             | 63    | +            | .2041 $\pm$ .885*  |
| II & III        | 1120 | 160  | 64388          | 1406           | 8883  | -            | .1680 $\pm$ .723*  |
| II & IV         | 1120 | 120  | 64388          | 776            | 6746  | +            | .0851 $\pm$ .362   |
| II & V          | 1120 | 120  | 64388          | 778            | 6682  | -            | .1222 $\pm$ .522*  |
| II & VI         | 1120 | 200  | 64388          | 2126           | 11401 | +            | .4384 $\pm$ 2.07*  |
| II & VII        | 1120 | 40   | 64388          | 102            | 2300  | +            | .3132 $\pm$ 1.40*  |
| II & VIII       | 1120 | 100  | 64388          | 550            | 5425  | -            | .6060 $\pm$ 3.23*  |
| II & IX         | 1120 | 60   | 64388          | 208            | 3390  | +            | .1388 $\pm$ .589*  |
| II & X          | 1120 | 20   | 64388          | 32             | 1131  | +            | .0777 $\pm$ .332   |
| III & IV        | 160  | 120  | 1406           | 776            | 968   | +            | .0952 $\pm$ .406   |
| III & V         | 160  | 120  | 1406           | 778            | 975   | +            | .1755 $\pm$ .756*  |
| III & VI        | 160  | 200  | 1406           | 2126           | 1539  | -            | .4841 $\pm$ 2.35*  |
| III & VII       | 160  | 40   | 1406           | 102            | 307   | -            | .2469 $\pm$ 1.08*  |
| III & VIII      | 160  | 100  | 1406           | 550            | 801   | +            | .0126 $\pm$ .054   |
| III & IX        | 160  | 60   | 1406           | 208            | 498   | +            | .3031 $\pm$ 1.35*  |
| III & X         | 160  | 20   | 1406           | 32             | 161   | +            | .0257 $\pm$ .109   |
| IV & V          | 120  | 120  | 776            | 778            | 738   | +            | .3158 $\pm$ 1.41*  |
| IV & VI         | 120  | 200  | 776            | 2126           | 1196  | -            | .0476 $\pm$ .202   |
| IV & VII        | 120  | 40   | 776            | 102            | 250   | +            | .2849 $\pm$ 1.26*  |
| IV & VIII       | 120  | 100  | 776            | 540            | 609   | +            | .1701 $\pm$ .732*  |
| IV & IX         | 120  | 60   | 776            | 208            | 375   | +            | .3788 $\pm$ 1.74*  |
| IV & X          | 120  | 20   | 776            | 32             | 121   | +            | .0386 $\pm$ 1.164  |
| V & VI          | 120  | 200  | 778            | 2126           | 1160  | -            | .9358 $\pm$ 11.26* |
| V & VII         | 120  | 40   | 778            | 102            | 241   | +            | .0280 $\pm$ .119   |
| V & VIII        | 120  | 100  | 778            | 550            | 647   | +            | .8728 $\pm$ 7.59*  |
| V & IX          | 120  | 60   | 778            | 208            | 379   | +            | .2769 $\pm$ 1.22*  |
| V & X           | 120  | 20   | 778            | 32             | 122   | +            | .0758 $\pm$ .323   |
| VI & VII        | 200  | 40   | 2126           | 102            | 408   | +            | .1519 $\pm$ .652*  |
| VI & VIII       | 200  | 100  | 2126           | 550            | 972   | -            | .3528 $\pm$ 1.60*  |
| VI & IX         | 200  | 60   | 2126           | 208            | 587   | -            | .2189 $\pm$ .952*  |
| VI & X          | 200  | 20   | 2126           | 32             | 195   | -            | .1286 $\pm$ .550*  |
| VII & VIII      | 40   | 100  | 102            | 550            | 206   | +            | .1809 $\pm$ .780*  |
| VII & IX        | 40   | 60   | 102            | 208            | 122   | +            | .0806 $\pm$ .343   |
| VII & X         | 40   | 20   | 102            | 32             | 34    | -            | .3693 $\pm$ 1.69*  |
| VIII & IX       | 100  | 60   | 550            | 208            | 302   | +            | .0535 $\pm$ .227   |
| VIII & X        | 100  | 20   | 550            | 32             | 96    | -            | .1633 $\pm$ .702*  |
| IX & X          | 60   | 20   | 208            | 32             | 62    | +            | .1091 $\pm$ .466*  |

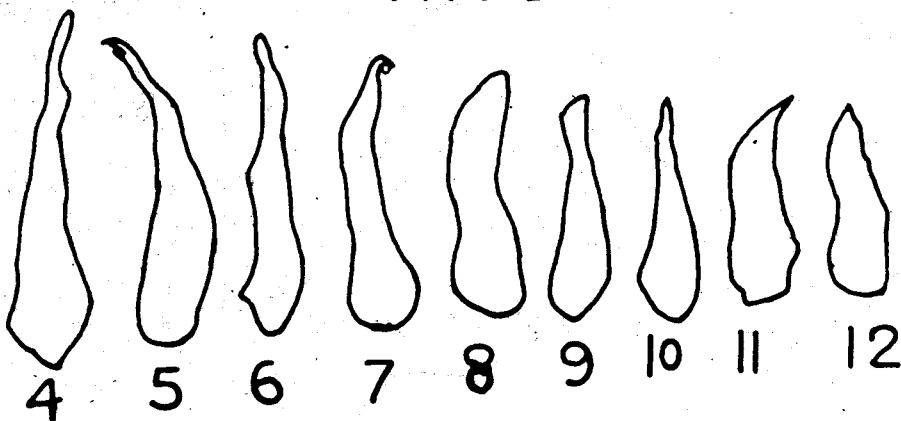
\* Significant (for  $n=20$ , significant value of ' $r$ ' = 0.423)



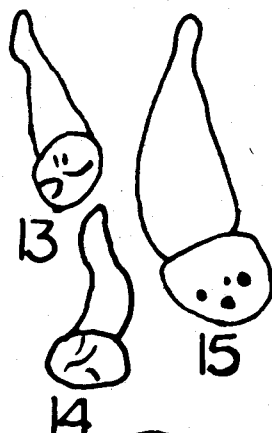
TYPE-I



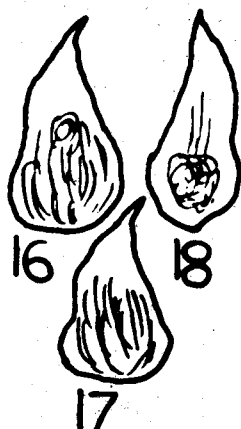
TYPE-II



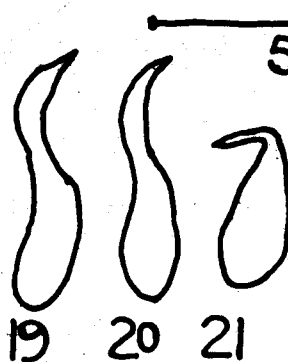
TYPE-III



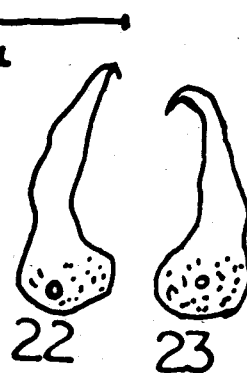
TYPE-IV



TYPE-V



TYPE-VI

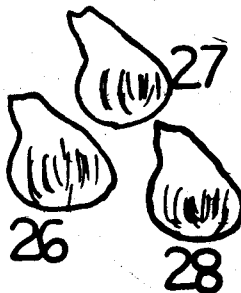


50  $\mu$

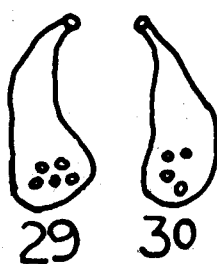
TYPE-VII



TYPE-VIII



TYPE-IX



TYPE-X

