

completed readily in the following manner. Figure 1A shows a fluorescence spectrum recorded with a detector that has the response characteristic shown in Fig. 1B. The true fluorescence spectrum (and hence true fluorescence maximum) is obtained by taking the product of 1A and 1B at each point and is illustrated by Fig. 1C. The true and apparent maxima are marked appropriately and differ by 7 m $\mu$ .

The naphthols provide an excellent illustration of this phenomenon. When a 1P28 photomultiplier tube is used, the visible fluorescence maximum of 1-naphthol, dissolved in concentrated sulfuric acid, is recorded at 528 m $\mu$ . The value obtained for the maximum, when corrected as described in the foregoing paragraph, is 544 m $\mu$ , a change of 16 m $\mu$ . Likewise, 1-naphthol dissolved in 0.2N sodium hydroxide shows an apparent maximum at 486 m $\mu$ , while the corrected maximum is at 500 m $\mu$ . Also, 2-naphthol dissolved in 0.2N sodium hydroxide and 0.1N sulfuric acid has apparent maxima of 424 m $\mu$  and 357 m $\mu$ , respectively, which, when corrected, give values of 429 m $\mu$  and 358 m $\mu$ . In general, response curves provided by the manufacturer of the phototube are sufficiently accurate, although for most precise results the detector may be calibrated by the National Bureau of Standards.

Scattered light from the source of excitation can also affect the fluorescence maximum and is dependent on the spectral band-pass of the analyzer monochrometer. If the fluorescence of the solution is weak, scattered light from the source can be reflected from the solution into the analyzer. If this light is of the appropriate wavelength (that is, if it is near the fluorescence maximum) it can cause a shift in the maximum recorded. The extent of this phenomenon can be readily checked by running blank solutions.

The accuracy of the wavelength calibration of the analyzer monochrometer is very important in locating the fluorescence maximum. The calibration can be checked with a low-pressure mercury arc or a hydrogen lamp (4).

Another factor is the slit width (spectral band-pass) of the analyzer monochrometer, for if this is too large it is conceivable that a false maximum might be obtained. This is especially true if the spectral band-pass changes with wavelength, as it does for prism instruments. The effect of this factor can be evaluated by running spectra at increasingly smaller slit widths and noting any shifts in the maximum.

A number of incidental factors can affect the fluorescence maximum. An impurity in the sample material or the solvent would have an effect if the impurity present were fluorescent. Many materials

(such as benzene or alcohol) may contain traces of fluorescent impurities which could cause errors of this sort. It is often difficult to remove impurities of this type by conventional purification procedures. Fluorescence that results from the solvent can be detected by use of a blank.

A number of factors have been presented in this discussion which influence the value for the fluorescence maximum of a given compound, recorded on a specific instrument. If recorded curves have not been corrected, they are specific only for that particular instrument. However, they may be reproducible from laboratory to laboratory on an instrument of a given type, but this is not a sufficient criterion for a true maximum. It is therefore proposed that these uncorrected maxima be called *apparent* fluorescence maxima and that only those which are corrected for the factors discussed in this report be labeled *true* fluorescence maxima.

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### Formation of Gibbsite Aggregates in Latosols Developed on Volcanic Ash

The segregation of gibbsite on dehydration into light-colored, irregular-shaped aggregates has been observed in certain Hawaiian soils which have developed on andesitic volcanic ash occurring in the rain forest areas. These soils have been classified by Cline *et al.* (1) into the hydrol humic latosol soil group. These soils have been developed by the intense and rapid weathering of andesitic volcanic ash under a warm climate having a heavy rainfall ranging from 120 to 350 inches with no season which can be considered dry. Under these conditions, the primary silicate minerals of the volcanic ash have decomposed, and many products of weathering have been leached from the soil. The free and rapid percolation of water has provided conditions favoring the removal of the released soluble silica. The desilication has

Table 1. Chemical composition of the light-colored aggregates of gibbsite formed on dehydration of soils of the hydrol humic latosol group. All figures are percentages.

Location	SiO <sub>2</sub>	Al <sub>2</sub> O <sub>3</sub>	Fe <sub>2</sub> O <sub>3</sub>	TiO <sub>2</sub>	H <sub>2</sub> O
Onomea	0.89	61.88	0.88	2.76	31.88
Kaiwiki	0.72	63.03	1.20	3.02	33.09
Hilo Sugar Co.	0.62	62.22	2.28	1.28	33.40
Hakalau	0.90	61.60	2.08	4.52	31.52
Pepeekeo	0.69	62.81	0.88	2.22	32.60
Hilo Forest Reserve	0.58	63.30	0.90	1.22	32.89
Average	0.72	62.46	1.37	2.50	32.55

been accelerated by the organic acids, which are provided by the decomposition of the mixed fern vegetation.

The greatest development of the soils of the hydrol humic latosol group occurs on the beds of andesitic ash found on the slopes of Mauna Kea on the island of Hawaii. These soils are smeary, gelatinous clays which, upon dehydration, form a mixture of light- and dark-colored aggregates. The aggregates will not rehydrate to form the clay but instead become very water-stable aggregates. The volume weight of these soils is extremely low, ranging from 0.1 to 0.7 and averaging approximately 0.5. The soils often contain from 60 to 65 percent water; however, they have formed a very stable land surface.

The mineral and chemical composition of typical soils of the hydrol humic latosol group has been reported by Tamura *et al.* (2). The major mineral constituents which were identified and estimated in this report (3) are as follows: gibbsite, 25 to 33 percent; allophane, 13 to 26 percent; goethite, 10 to 34 percent; magnetite, 6 to 19 percent; and anatase, mica, silica, and quartz in minor amounts.

On drying, these soils lose volume. In the process of drying, light reddish-brown mottles are formed by congealing gel-like structures which separate from the darker-colored matrix. On further drying, the light-colored areas form solid aggregates which feel gritty in the dehydrating system. Finally, on complete dehydration, a mixture of light- and dark-colored aggregates is formed. The light-colored aggregates have been identified as gibbsite, the trihydrate of aluminum oxide, by chemical and differential thermal analysis. Tamura (4) has also confirmed this identification by x-ray diffraction procedures. The chemical composition of the six separations of light-colored aggregates is given in Table 1. The average alumina content of the aggregates is 62.46 percent, and the average water loss between 110° and 400°C is 32.55 percent. Only minor quantities of silica, iron oxide, and titanium oxide were found in these aggregates.

The dark-colored aggregates have a high content of silica, more than 20 percent, and also iron oxide, 30 to 40 percent. These aggregates are strongly magnetic, thus providing a method of separation from the light-colored aggregates.

The process by which the gibbsite aggregates are formed has not been established. The indications strongly suggest that the alumina has been precipitated from the percolating waters in the pores, cavities, and drainage channels occurring in the soil. The best evidence of this is the fibrous structures often found in these aggregates. The segregation of gibbsite into discrete aggregates may provide a means for its commercial development as a source of alumina.

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#### References and Notes

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### Preparation and Properties of Antihuman Properdin Rabbit Serum

A specific antiserum for properdin is desirable (i) as a tool for characterizing properdin and its interactions and (ii) for the possible development of quantitative immunochemical procedures for the measurement of properdin. In addition, studies on the antigenicity of properdin are necessary to determine whether heterologous properdins can be employed for *in vivo* experiments.

Partially purified properdin (1000 units/mg of protein nitrogen) was prepared from human serum as is described elsewhere (1) and was measured by the zymosan assay (1).

Albino rabbits were given six to ten 1000-unit injections of human properdin intravenously over a period of 10 to 14 days. One to 3 months later they were given a single injection of 1000 units and were bled 2 to 4 days following this single injection. About 20 percent of the rabbits produced satisfactory antisera.

Serums were separated in the usual manner and were stored at  $-70^{\circ}\text{C}$ . Aliquots of anti-HP were heated at  $56^{\circ}\text{C}$  for 1 hour (anti-HP $\Delta$ ). Other aliquots of anti-HP $\Delta$  were incubated with 1 to 2 vol of human serum deficient in properdin (RP) for 1 hour at  $37^{\circ}\text{C}$  and then at  $1^{\circ}\text{C}$  overnight. The mixtures were centrifuged at 4000 rev/min for 1 hour at  $1^{\circ}\text{C}$ , and the supernatants [anti-

HP(A)] were stored at  $-70^{\circ}\text{C}$  until used. Normal rabbit serum, antihuman gamma globulin rabbit serum and antihuman serum rabbit serum (Coombs) were treated in a similar fashion.

For the measurement of antiproperdin activity, 0.2 ml of varying dilutions of antisera were added to known amounts of either purified properdin or human serum of known properdin content. The mixtures were incubated at  $37^{\circ}\text{C}$  for 1 hour and, in certain experiments, at  $1^{\circ}\text{C}$  overnight. The samples were then centrifuged at 4000 rev/min for 1 hour, and the supernatants were titered for properdin and components of complement. Adequate controls, which included normal rabbit serum, Coombs serum, and isotonic sodium chloride or buffer, were always run simultaneously. The antibody titers are expressed as the number of units of properdin inactivated or neutralized by 1 ml of antiserum. Thus, if 0.2 ml of a 1/5 dilution of antiserum inactivated 5 units of properdin, the sample was considered to contain antibodies capable of neutralizing 125 units of properdin per milliliter of serum.

The results of numerous experiments on the serological properties of anti-HP are summarized in Table 1. It will be seen that the antibody to properdin was stable to heating at  $56^{\circ}\text{C}$  for 1 hour. Anti-HP and anti-HP $\Delta$  also formed precipitates with fresh human serum and inactivated complement. However, adsorption of anti-HP $\Delta$  with RP (human serum deficient in properdin) removed all demonstrable complement-fixing and precipitating antibodies and provided a serum [anti-HP(A)] which appears by present criteria to inactivate properdin

specifically. Preliminary experiments indicate that purified properdins from other species are also inactivated to varying degrees by anti-HP(A). Normal rabbit serum and several Coombs serums did not inactivate properdin.

Table 1 shows that treatment of anti-HP(A) with purified properdin or properdin-zymosan complexes (PZ) completely removes the antibody to properdin. Complexes formed from zymosan and heated serum, as well as zymosan alone, were ineffective. The specificity of properdin for the removal of antibody from anti-HP(A) and the lack of dependence on temperature for this removal further substantiate the concept that the substance in immune serum which inactivates properdin is a specific antibody to properdin.

The results presented so far were obtained by measuring properdin in the zymosan assay. It is known that the properdin system inactivates certain viruses (2) and phages (3), kills certain bacteria (4) and protozoans (5), and hemolyzes abnormal red cells (6). It seemed necessary to determine whether the anti-HP(A) employed in this study would also inactivate properdin in these systems. Accordingly, samples of anti-HP(A) were sent to the investigators mentioned in Table 2 for evaluation of the activities of this antiserum. Their results are summarized in Table 2, which shows that these investigators employing different methods of assay for the measurement of properdin arrived at similar values for the antiproperdin potency of the anti-HP(A).

The foregoing experiments (7) show that immunization of rabbits with puri-

Table 1. The inactivation of human properdin by antihuman properdin rabbit serum.

Sample	Units of properdin inactivated per milliliter of serum	Complement-fixation with human serum	Precipitation with human serum
Untreated antiproperdin rabbit serum (anti-HP)	200-300	+++	+++
Anti-HP heated at $56^{\circ}\text{C}$ for 1 hour (anti-HP $\Delta$ )	200-300	+++	+++
Anti-HP $\Delta$ adsorbed with RP [anti-HP(A)]*	80-120	-	-
Anti-HP(A) adsorbed with properdin†	0	-	-
Anti-HP(A) adsorbed with properdin-zymosan complex‡	0	-	-
Anti-HP(A) adsorbed with zymosan	80-120	-	-
Antihuman $\gamma$ -globulin or antihuman serum rabbit serums§	0	$\pm$ to +++	$\pm$ to +++
Normal rabbit serum	0	-	-

\* 1.5 ml of RP (human serum deficient in properdin) per milliliter of anti-HP $\Delta$ .

† 100 units of purified human properdin per milliliter of anti-HP(A).

‡ Complex was formed from 20 mg of zymosan and 10 ml of fresh human serum. Complexes formed from heated serum were ineffective in adsorbing antiproperdin.

§ Different Coombs serums varied in their ability to fix complement or to form immune precipitates.