Table 1. Statistical analysis of data obtained from phosphatase activity in the whole salivas of 26 caries-free and 26 caries-active persons.

Activity	Caries-free	Caries-active
*Alkaline**		
Range	0.06– 1.98	0.06– 1.98
Mean ($\bar{x}$)	0.58	0.68
S. D. (σ)	0.66	0.50
	$\frac{\text{Dev.}}{\sigma} = \frac{\bar{x}_1 - \bar{x}_2 - O}{\sigma x_1 - x_2} = 0.61$	
*Acid**		
Range	0.90–12.60	2.40–13.80
Mean ($\bar{x}$)	5.65	7.61
S. D. (σ)	4.11	3.30
	$\frac{\text{Dev.}}{\sigma} = \frac{\bar{x}_1 - \bar{x}_2 - C}{\sigma \bar{x}_1 - \bar{x}_2} = 1.9$	

* Units of phosphatase per 100 ml of whole saliva.

caries activity and phosphatases found in whole stimulated saliva. Analyses for both acid and alkaline phosphatase were performed on a randomly selected group and on caries-free and caries-active individuals.

Fifteen milliliters of paraffin-stimulated saliva were collected from each of 100 naval recruits ranging in age from 17 to 20 years. Acute cases of gingival infection were eliminated from this otherwise randomly selected group. Similar saliva samples were collected from 26 caries-free and 26 caries-active individuals who had had no dental restorations. The caries-free group was selected by clinical and radiographic examination. The caries-active individuals were selected clinically on the basis of the existence of ten or more carious lesions.

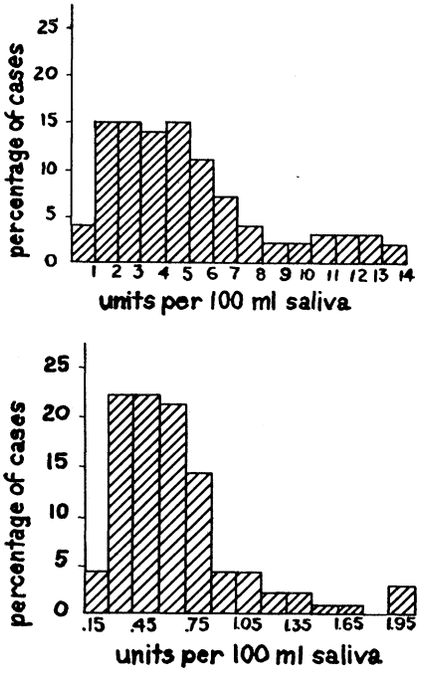

Fig. 1. Phosphatase activity in whole salivas of 100 randomly selected naval personnel of age ranging from 17 to 20 years. (Top) Acid pohsphatase: mean, 4.72; range, 0.60 to 13.8; standard deviation, ± 3. (Bottom) Alkaline phosphatase: mean, 0.57; range, 0.12 to 1.92; standard deviation, ± 0.405.

The colorimetric method of Seligman *et al.* (*10*), as adapted for saliva tests by Chauncey (*3*), was used for phosphatase determinations. The Coleman junior spectrophotometer at an optimum wavelength of 525 mμ and 10 by 75-mm cuvettes were used.

The mean acid phosphatase activity in whole salivas of 100 randomly selected persons was 4.72 units/100 ml, while that for alkaline phosphatase was 0.57 units/100 ml (Fig. 1). The mean acid and alkaline phosphatase activities in whole salivas of 26 caries-free individuals were 5.65 units/100 ml and 0.58 units/100 ml, respectively. The caries-active group showed mean phosphatase activities of 7.61 units/100 ml (acid) and 0.68 units/100 ml (alkaline).

Statistical analysis of the differences in the mean acid and alkaline phosphatase levels for caries-free and caries-active groups did not reveal significance. The probability of obtaining the observed differences in the means for acid phosphatase was 7 percent or less; for alkaline phosphatase, it was 54 percent or less (Table 1).

That oral phosphatases are predominately of bacterial cell origin seems well established (*1–4*). Rosebury (*6*) and others (*4, 7*) have ascribed possible roles to phosphatases in saliva that are based solely on known activity characteristics of these enzymes isolated from other tissues (bone, liver, kidney, serum, and so forth). The mere existence of a phosphatase associated with the bacterial cell does not, however, preclude the action of this enzyme according to previously accepted theory. Thus it becomes important to look on the oral phosphatases as entities and to characterize them according to the specific oral debility in which they are suspected to be taking part. For instance, recent tests in this laboratory showed that acid phosphatase of the parotid secretion was inhibited by tartrate, which has been shown to inhibit prostatic phosphatase (*11*). This would immediately bring to mind specific correlations regarding tests for cancer of prostate gland. However, characterizations of both parotid and prostatic enzymes become essential before conclusions may be drawn.

The failure to correlate the presence of phosphatases in saliva with certain oral debilities (dental caries, calculus formation, periodontal disease, and so forth) probably results from the fact that these enzymes are present in so many of the organisms normally present in the mouth that any association is obscured.

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25 August 1955

Possible Function of Serum Proteins in Tissue Culture

In a recent article by Eagle (*1*) it was found that, with the exception of the necessity of serum proteins, completely defined chemical media are possible that support both growth and multiplication in certain lines of mammalian cells. The possibility that the serum proteins contribute as yet undetected trace elements or vitamins is being investigated by him (*1*, p. 503).

Although the presence of such trace elements or vitamins is certainly a possibility, there is another alternative that I feel should be considered. Both from the work described and what is known about protein chemistry and function, it seems that the essential substances could be proteins, perhaps certain of the serum proteins themselves.

From the results given in the paper (*1*, p. 503) it appears that the activities of the protein fractions were determined by the extent to which denaturation could not occur in the fractionation procedure. Exhaustive dialysis would remove most of the salts and other impurities, and this is known to render many proteins unstable and susceptible to structural changes (*2*, pp. 211–213; *3*). Alcohol is a denaturing agent for many proteins (*2*, p. 207; *4*, p. 173), and

there is evidence that serum proteins resulting from alcohol fractionation are denatured to the extent that, although still soluble, they are poorly metabolized when they are injected into an animal (*4*, pp. 460–461). Only the salting out method by the use of neutral salts is thought to leave the protein fractions in their native, unaltered form (*4*, p. 173) and apparently only by this method was Eagle able to obtain fractions that were consistent in their activity.

The possibility that specific proteins are essential for the activities of at least some mammalian cells would also seem compatible with some of the properties and roles that have been described for certain proteins. They could act on the cell membrane and affect its permeability to other materials. There is evidence that the protein hormone insulin acts in this way (*5*). There is good evidence that certain plasma proteins can enter cells fairly readily (*6*); and, once inside the cell, the essential protein may act by complexing smaller compounds whose activity may depend on this action. The tremendous importance of this property of proteins and the dependance of the biological function of many compounds on the alteration of properties resulting from such complex formation has been pointed out in detail by Needham (*7*). It seems to be an accepted fact that protein hormones somehow alter the metabolism of the cells they affect; and, because some proteins are present in relative abundance, this should not in itself rule out an essential metabolic role for them.

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22 November 1955

Early Man and Fossil Bison

At certain sites in and near the Great Plains, there are associations of fossil bison and early types of projectile points. Correlating these associations, I have developed a possible sequence of certain projectile points and of contemporaneous fossil bison forms (Fig. 1) (*1*).

In 1947, Skinner and Kaisen enumerated the known localities where fossil bison are represented (*2*). At a number of these localities, Folsom, Plainview, and Scottsbluff points have been found. Since 1947, Scottsbluff points have also been identified at the Finley site in association with *Bison* (*Bison*) *occidentalis* (*3*).

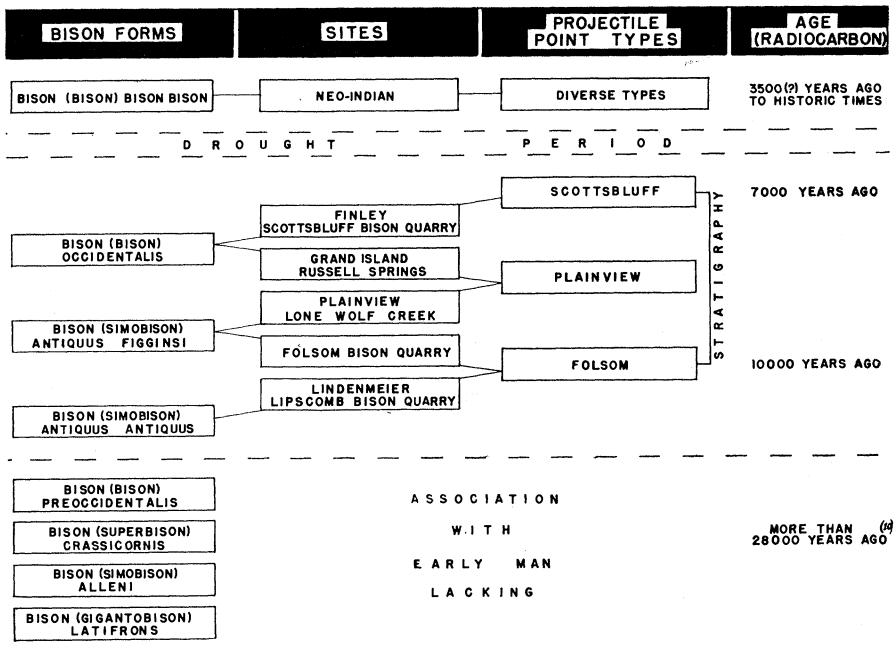

Fig. 1. Associations of bison forms with sites and projectile point types in the Great Plains.

Excavations at the MacHaffie site revealed that the Folsom complex is older than the Scottsbluff (*4*). This fact has been confirmed elsewhere by radiocarbon dates on a Folsom site (9883 ± 350 years ago) and on a Scottsbluff site (6876 ± 250 years ago)—dates that indicate an age difference of some 3000 years (*5*).

The Folsom-Scottsbluff succession having been established, it is evident that Fig. 1 shows the best possible way of correlating the fossil bison and projectile points.

The number of sites with associations will have to be increased before the results of the correlation can be regarded as established fact. Nevertheless, until contradictory evidence comes to light, it provides a working hypothesis.

Sound contradictory evidence is lacking. At the Lime Creek site, a Scottsbluff layer was reportedly found stratigraphically below a layer attributed to the Plainview complex (*6*). However, according to Krieger (who originally conceptualized the Plainview point), the Lime Creek specimens are not Plainview (*7*).

Plainview points may prove to be younger than Scottsbluff on the basis of a supposed geologic relationship between the Lime Creek site, with its Scottsbluff layer, and the nearby Red Smoke site, where authentic Plainview points were found. The geologic evidence, however, has not yet been published in a convincing manner.

One potential flaw in the chain of evidence must be noted. The points from Grand Island and Russell Springs are not typical Plainview points. Probably they are merely variants of the Plainview type. At the Red Smoke site, these so-called Meserve points were found in the same cultural layer as typical Plainview points.

Early man (Paleo-Indian) occupations appear to be separated from the later (Neo-Indian) occupations of the plains by a drouth period of some 3000 years, beginning about 6500 to 7000 years ago. Fossil forms of bison have not yet been found in any of the known Neo-Indian sites. And, with one possible exception, modern bison have not been found in sites attributed to early man on the Great Plains.

The possible exception is the Agate Basin site in Wyoming (*8*). There, the bison (said to be of historic species) were not classified according to the system Skinner and Kaisen developed. Unfortunately, they were discarded from the U.S. National Museum because they appeared to be only a superfluous addition to an already adequate collection of modern bison (*9*).

A renewed excavation of this site and a reexamination of its bison remains will show whether or not the historic species lived contemporaneously with *occidentalis* in the Paleo-Indian period. Contemporaneity seems to be out of the question, because the historic plains