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Note

Improved determination of plasma phosphoserine

ANGELO MONDINO, SILVANO FUMERO and GIANNI BONGIOVANNI
Istituto di Ricerche Biomediche Antoine Marxer S.p.A., R.B.M., 10015 Ivrea (Italy)
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The displacement of tryptophan from plasma albumin by means of sodium dodecylsulphate (SDS) has been reported in a preceding paper[1]. In fact, if the plasma sample is treated with a 0.5% aqueous solution of SDS before deproteinization with sulphosalicylic acid (SSA), the recovery of tryptophan becomes complete once the deproteinized sample is submitted to ion-exchange chromatography. In this way, the need for an extra analysis, usually fluorimetric[2], is eliminated.

In the light of these findings, in our laboratories the amino acid spectrum of human plasma is usually determined after treatment of the samples by this method. It has been observed that the phosphoserine peak, which formerly was usually very small (if not completely absent), increased dramatically.

It could be assumed that phosphoserine, as well as tryptophan, is transported by plasma proteins, from which the SSA deproteinization was capable of liberating it only in a very small amount, whereas plasma treatment with SDS, prior to protein precipitation, permits the liberation of phosphoserine.

Table I reports results obtained by analyzing six human plasma samples both after simple deproteinization with an SSA solution, as described earlier[3], and after treatment with a freshly prepared 0.5% aqueous solution of SDS followed by protein precipitation with SSA according to the procedure described earlier[1].

The amino acid chromatography, the apparatus and methods employed are the same as those described earlier[1,4,5]. It can be seen by comparing the results in column A with those in column B, in Table I that the values found for the phospho-

TABLE I

PHOSPHOSERINE LEVELS IN HUMAN PLASMA

Sample No.	*A**	*B***
1	0.10	0.59
2	0.13	1.23
3	0.06	0.58
4	0.09	0.70
5	0.10	0.50
6	0.27	1.40

* A = plasma deproteinized with 3.75% SSA only.
** B = plasma treated with 0.5% SDS and then deproteinized with 5% SSA.

serine plasma levels with the method described are, on average, about seven times higher than before. It can also be seen that SSA, when used without pre-treatment with SDS, is only capable of liberating an amount of phosphoserine ranging from 10 to 20% of the amount freed under the displacing action of SDS. It should be recalled that the SSA deproteinization was capable of liberating about 94% of the amount of tryptophan that was displaced by pre-treatment with SDS.

It is well known[1,6] that tryptophan is bound to albumin by its carboxylic group, whose pK value is presumably higher than the plasma pH after addition of SSA. This could be the explanation of the higher tryptophan recovery compared with the recovery of phosphoserine, which is presumably bound by its hydroxylic groups, having a lower pK. Consequently, it can be postulated that phosphoserine is not bound to albumin (like tryptophan), but rather to some protein fraction having more basic properties, such as globulins. Further research is advisable in order to clarify how this amino acid is transported in plasma, and which proteins are responsible.

In conclusion, the improved determination of this compound achievable with the present method of deproteinization can be recommended as it can contribute new information on its metabolic significance.

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