THE METHOD FOR PRODUCTION OF HIGH PURITY CARRIER FREE ORTOPHOSPHORIC ACID LABELED WITH ISOTOPES PHOSPHORUS-32 AND PHOSPHORUS-33.

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Extensive application for various radioactive isotopes was found in an extremity of the 20-th century in a science and production. Labeled compounds are used with growing effectiveness in a molecular biology, gene engineering, medicine and other areas. Phosphorus-32 and Phosphorus-33 isotopes as a different labeled compounds that are used mainly in molecular biology are produced at the "Radiopreparat" enterprise of the Institute of Nuclear Physics of Academy of Sciences of Uzbekistan Republic. The quality of labeled preparations is very high. The specifications for above mentioned preparations corresponds to demands most of customers in different countries. P-32 or P-33 labeled orthophosphoric acid has high radiochemical purity (more than 99 %) and specific radioactivity close to theoretical.

A lot of different methods are known for preparing of orthophosphoric acid (H₃³²P₀₄ and H₃³³P₀₄ from a neutrons irradiated sulfur. In one of described methods the irradiated sulfur is transferring to a quartz distillation vessel connected with the cooled receiver which is connected to a vacuum system in turn. A sublimation of sulfur is effected at 180℃ and vacuum about 1 mm of mercury. After the completion of sublimation the quartz distillation vessel is cooled and filled up with 0,1 M HCl containing Hydrogen Peroxyde. The solution is boiled within 2 hours after the vessel is connected to a return refrigerator. The prepared cooled solution is passed through a column packed with the cationexchange resin "DOWEX-50" to remove a cation impurities. The purified solution is evaporated after adding Hydrogen Peroxyde for destruction of organic compounds.

Orthophosphoric acid prepared by the described above method has radiochemical purity about 95 % and output of the target product 99%.

In the other work the technique for preparing of orthophosphoric acid is based on the ability of the irradiated sulfur to be dissolved in a paraffin. Radioactive phosphorus as orthophosphoric acid should be then be extracted by 0,1 M Hydrochloric acid.

These and other methods for preparing of ³²P and ³³P orthophosphoric acid have the defects that are not desirable when Sulfur-33 is used as raw material for ³³P production.
As result of long-term practical experience the effective method had been developed at the enterprise "Radiopreparat" for preparing of $^{32}\text{P}$ and $^{33}\text{P}$ orthophosphoric which allow to receive the preparations of high quality with the specific radioactivity close to theoretical.

Especially it concerns the method of $^{33}\text{P}$ orthophosphoric acid production. In connection with $^{33}\text{S}$ very high cost there was a problem to decrease of raw material losses during processing simultaneously with the purpose of its repeated secondary use.

To achieve maximum output of $^{33}\text{P}$ radioactivity per gram of Sulfur-33 or Sulfur-32 the most important significance have the conditions of the targets irradiation. The dependence of an output of radioactivity from irradiation time and from of neutrons flux is shown on picture 1 and picture 2.

**Figure 1.** The dependence of P-32 radioactivity output depending on the irradiation time and neutrons flux.

**Figure 2.** The dependence of P-33 radioactivity output depending on the irradiation time and neutrons flux.
In the outcome of long-term experience we came to a conclusion that the optimum time of irradiation for $^{33}\text{S}$ is about 500 hours. If the irradiation exposure is more than 500 hours high percentage of quartz ampoules destruction is happened and high lost of Sulfur-33 take place. This is one of the problems we collide periodically. Though in this direction is made a lot we proceed with the experiments. In particular to avoid high loss of raw material when destructions of the ampoules take place during irradiation we started to seal the ampoules containing Sulfur into aluminium tubes before the irradiation. It has allowed us to save expensive raw material and avoid cooling water contamination in the reactor. To eliminate overpressure in the quartz ampoules with Sulfur-33 during irradiation which will be derivated from a radiation heating we started to remove air to create vacuum. As result of these measures the postirradiation amount of broken quartz ampoules were decreased considerably. But as it was marked above the destruction of the ampoules is happened periodically and we proceed the investigation works in this direction.

The points of the method do not differ very much from the known "classical" with the difference that the quartz device created and patented "RADIOPREPARAT" enterprise is applied for the $^{33}\text{S}$ sublimation and that into technological process are included additional stage of purification of the intermediate solution from the mechanical impurities and developed the method of a maximum desorption of $^{33}\text{P}$ before "DOWEX" column chromatography approaches (see the Figure 3).

**Figure 3.** Technological scheme for P-32 and P-33 Orthophosphoric Asid production.
The irradiated ampoule containing $^{33}\text{S}$ is put into quartz device. Sulfur is sublimating at the temperature approximately 120°C and vacuum about 0.02 mm of mercury. After the sublimation the sulfur is condensed on the cooled part of the quartz tube. Upon termination of a sublimation a quartz tube is cut off. After appropriate preparation this part of the tube is sealed under vacuum from the both sides with the receiving the new ampoule ready for a repeated irradiation.

The technology for preparing of orthophosphoric acid includes the stages of phosphate ions desorption from the walls of the quartz ampoule by means of Hydrochloric acid solution boiling, filtration of an obtained solution, disposal of Hydrochloric acid solution by repeating evaporation with adding of $\text{H}_2\text{O}$, keeping of the solution at room temperature to mature, the application to the columns with cation- and anion-exchange resins and the elution of Orthophosphoric acid by Hydrochloric acid solution.

Keeping of the solution at room temperature before the chromatography is necessary for a maximum desorption of Orthophosphoric acid from walls of the ampoule. Experimentally it was established that keeping of the solution not less than one hour ensures desorption not less than 95%.

The parameters of the Orthophosphoric acid preparations are demonstrated by the following data:

$^{32}\text{P}$/Orthophosphoric acid carrier free, in 0.04 M HCl,

- specific radioactivity - 8500-9000 Ci/mmol,
- volume radioactivity - 100-2000 mCi/ml,
- radiochemical purity - more than 99% as orthophosphate,
- radionuclidic purity - more than 99%,
- biological activity - not less than 95% in 10 minutes
- enzymatic conversion from ADP to $\gamma^{32}\text{P}/\text{ATP}$.

$^{33}\text{P}$/orthophosphoric acid, carrier free, in 0.04 M HCl or in water solution,

- specific radioactivity - 4500-5000 Ci/mmol,
- volume radioactivity - 500-2000 mCi/ml,
- radiochemical purity - more than 99% as orthophosphate,
- radionuclidic purity - more than 99% ($^{32}\text{P}$ less than 0.5%),
- biological activity - not less than 95% in 10 minutes
- enzymatic conversion from ADP to $\gamma^{33}\text{P}/\text{ATP}$.