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PROCESS FOR STERILIZATION OF ORGANIC SUBSTANCES CONTAINING MICROORGANISMES, BACTERIA, FUNGI OR SPORES

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The present invention relates to a process for sterilization or disinfection of any organic substances, or substances with organic constituents containing microorganismes, bacteria, fungi or spores, which substances not per se offer suitable possibilities for spore germination, such as guts for catguts, hair or bristles for brushes, fibrous material of any kind and shape, articles of rubber and silk, for instance for use in surgery, any material which has to be preserved, such as hormons, vitamins and medicines, any material which has to be delivered for germs before further manufacturing or use, such as drugs or tobacco leaves and other substances, wholly or partly of artificial, vegetable or animal origin, for instance products of animal organs, whether the substances be in solid or liquid or semiliquid state.

The processes commonly used heretofore for sterilization of material containing or contaminated with spores, for instance guts for catgut, suffer from the drawback that no complete sterilization can be attained with security, but only a certain disinfection, as the resistant microbic spores will not be killed, for instance spores of the bacteria of anthrax, tetanus and gas-gangrene. Experience has thus shown that commercial catgut threads, disinfected by heating, at various stages of the manufacture, maybe repeatedly to 120° or 150° C. in organic liquids and the like, or having been treated in some disinfecting liquid, for instance a solution of iodine, contain a multitude of resistant spore-producing bactefia which similarly will not be killed by any further disinfection performed by the surgeon, before the use. Further, the quality of the organic substances will suffer greatly by the treatments, especially thermal treatments, to which they are exposed. Thus for instance the catgut acquires a relatively low tensile strength, for which reason it will easily burst during the use. The said drawbacks are remedied by the present process.

According to this process, the organic material, for instance the raw gut material for the manufacture of a catgut, is treated in such a manner that any spores of the sporeproducing microbes may be able to germinate, and the cultivation is performed during such a short time that the spores will certainly germinate, but the microbes will not have time to form new resistant spores, after which the microbes are killed by the subsequent exposure of the material to a disinfecting or sterilizing process which owing to the state into which the microbes have now been brought does not have for its object to kill the resistant spores. As the question is merely to kill the vegetative microbe forms that have less resistance to sterilizing than the spore form, it will be easily feasible, by a suitable method, to attain the result that the sterilization becomes complete, at the same time as the material will not be affected to any considerable extent.

It has previously been attempted to attain a more reliable catgut sterilization by cultivating previously the gut material in a culture medium, according to the German Patents No. 600,512 and No. 642,968. But the cultivations concerned have been of long duration, at least 4 and 10 days, respectively, during which there are ample possibilities for a production of spores that will not be killed more easily by the disinfection following, for which reason the sterilization attained in this manner will hardly be more reliable than without the preliminary cultivation, and in any case no sufficiently effective sterilization will be attained. The material will further be weakened at the same time during the lengthy action of the culture medium and the microbes growing in the same and causing for instance the production of alkaline metabolic substances that the first mentioned patent has for its object to neutralize by the addition of a source of acid. None of the two patents, however, is kept in force.

The decisive feature, according to the invention, is to select the conditions of cultivation in such a manner that all the spores of deleterious microbes at hand, aërobe as well as anaërobe ones, will germinate during the course of a relatively short time within which no new resistant forms of spores will be produced, and without the goods having been altered materially by the action of microbes. The maximum duration of the cultivation process will be about 72 hours or less, e. g. 24 hours or less. In a preferred execution of the process only a duration of a few hours is used, e. g. less than one to four or six hours, by use of liver broth or other media or substances which are able to accelerate spore germination.

The treatment may be performed, maybe after a requisite preliminary treatment of the material, by placing the same in a cultivating apparatus, at suitable and maybe alternating temperatures, and with a suitable culture medium, or with several such, containing perhaps oxygen-binding agents. These agents may be organic or inorganic chemicals or organic matters such as pieces of tissues. The cultivation may be performed in alternating atmospheres, under aërobe and anaërobe conditions—for instance under hydrogen, carbon diox-

ide, nitrogen, oxygen etc., or mixtures of such gases, or in vacuum. Then follows a treatment with microbicidal agents, for instance chemicals or ultra-violet rays, or special forms of heat-treatment, by which all of the microbes, including also the germinated spores, will be killed. This treatment may be performed after the material has been freed from the culture medium. A thermic treatment may for instance be performed on the quickly dried material. Owing to the quick drying, the spore-producing bacteria will produce no, or merely a few, weakened and only faintly heat-resistent spores which will be easily killed by the thermal treatment. It may often be opportune to alter the pH-value during the cultivation.

In certain cases the material may be of such a nature that for cultivation it cannot be placed, or does not have to be placed, in a special culture medium. By an addition of suitable substances, an alteration of the pH-value, or some other treatment, the material itself may be caused to offer suitable conditions for spore-germination and act as if it was a culture medium itself.

In order to be able to work with the greatest possible security, it may be desirable in certain cases, that the material should be subjected two or more times to the treatment for spore-germination and a subsequent sterilizing process. If chemicals are used for the sterilization, the same must first be eliminated e. g. neutralized, annihilated or removed to such an extent that a repeated cultivation can take place. In the repeated processes of cultivation and sterilization, the manners of treatment used may perhaps be modified, for instance by cultivating for longer periods, in other media et. cet.

For sterilization of catgut, the process mentioned in the example with one single cultivation and subsequent sterilization has proved to result in the complete killing of all microbes occurring in the initial material, since extensive controlling cultivations in various especially favourable media under aërobe conditions, as well as under various anaërobe conditions, during periods of up to one month, have not been able to demonstrate the growth of microbes of any kind.

If the process is used at an early stage of the manufacture of the goods concerned—like in the example as far as the catgut is concerned—the further treatment of the goods must be effected in a sterile, or mainly sterile manner, and in the last mentioned case it must be finished with a supplementary sterilization which nevertheless does not have to fill very rigid requirements, as it will be easy to make sure, that the material will merely be infected or contaminated with air bacteria and the like producing no spores and being easy to kill.

Thus the catgut threads, sterilized according to the process and afterwards treated in a merely partially asceptic way, have been able to be sterilized at last in such a lenient manner that the tensile strength of the finished goods will be from 50 to 100 p. Ct. higher than the tensile strength of the ordinary catgut threads. The process provides in this way, besides a securely sterile product, also the manufacture of a very strong catgut. Besides the now commonly used sheep and lamb guts, we may also use cheaper resorbable kinds of guts, for instance swine guts which earlier could not be used, because they would be weakened too much by the sterilization methods used heretofore, or guts may be used that heretofore would have to be rejected on account of a too high content of bacteria.

The process may be used to advantage for sterilization of for instance bristles, hair and fibrous substances of any kind and shape in raw and finished state. In this connection it may be mentioned that it has been very difficult to sterilize effectively animal bristles and hair from animals suffering from anthrax. In several cases diseases and deaths have therefore occurred owing to the use of unsufficiently disinfected bristles and hair, for instance shaving brushes. Among other uses of the process valuable in practices, we may also mention the sterilization of products gained from animal organs or from vegetable initial material, for instance hormon preparations and canned food. The sterilization may preferably be affected in a manner that is lenient to the goods.

Example

Sheep guts are treated preliminarily according to methods known per se, until the guts by scraping are freed from superfluous material, and they are cut into suitable lengths, to the ends of which the usual string straps are attached. Then they are placed into cultivation glasses with freshly boiled liver broth with pH of 7,6, in such a manner that the entire material is covered with the broth. Then the cultivation glass is placed in a Zeissler anaërobic jar which is pumped out to vacuum by means of an oil pump, and the glass is then placed in a thermostat at 37°C. for three hours. Then the cultivation glass is removed from the Zeissler jar, and is now left under aërobe conditions in a thermostat at 37°C. for one to two hours. Then the material is transferred to a sterile 1 p.Ct. solution of iodine in a potassium iodide solution, 3 in 200, poured into a sterile glass-bulb, in which it is treated for 24 to 48 hours. After this treatment, the material is transferred, in sterile manner, to a bulb with a sterile 10 p.Ct. solution of sodium thiosulphate in sodium carbonate solution, 1 in 100, and is treated here for 24 hours, in such a manner that the main part of the iodine is removed from the material. The latter is then transferred to a vessel with sterile water for washing. The result of the sterilization may be controlled by repeated cultivation, this time for longer periods.

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