

AMOXICILLIN TRIHYDRATE

5 The present invention relates to a product of amoxicillin trihydrate.

Water in β -lactam antibiotics in solid form may be present in different forms. Water may for instance be present as crystal water. Crystal water refers to water which is integrated in the molecular structure of the β -lactam antibiotic. The amount of crystal water in amoxicillin trihydrate containing 3 molecules of crystal water per molecule of
10 amoxicillin corresponds to about 12.9% crystal water. Free water refers to water that is available for exchange with the atmosphere. The amount of free water does not include the amount of water that is present as crystal water.

The free water content of a sample of a β -lactam antibiotic and the relative humidity of the air that is in contact with the sample influence each other. When a
15 sample of a β -lactam antibiotic is brought in contact with air, exchange of water between said sample and the air will generally take place until an equilibrium situation is established. In the equilibrium situation, the net exchange of water between the sample and the air that is in contact with the sample is zero. The free water content of a sample of a β -lactam antibiotic is typically given for a certain value of the relative humidity in the
20 abovementioned equilibrium situation at a given temperature. This value for the relative humidity is also referred to as equilibrium relative humidity.

The free water content of a sample can be determined by dynamic vapour sorption. The underlying principle is that the weight of the sample is monitored while it is contacted with air having a preconditioned relative humidity. As a result of water take-up
25 or water release the sample weight will change until the equilibrium situation has been established. The sample weight in this equilibrium situation is the sample weight corresponding to the equilibrium relative humidity, the latter being the relative humidity of the preconditioned air with which the sample is contacted. By repeating this procedure for different values of the equilibrium relative humidity, the sample weight can
30 be determined as a function of the equilibrium relative humidity. The free water content of a sample at temperature T can given by $((w_{ERH}-w_{ref})/w_{ref}) \cdot 100\%$, wherein w_{ERH} = sample weight, corresponding to equilibrium relative humidity ERH at temperature T and w_{ref} = sample weight, corresponding to a reference value for the

equilibrium relative humidity at temperature T, the reference value been chosen such that the free water content at said equilibrium relative humidity is close to zero.

In the application of a β -lactam antibiotic, the presence of free water may be a concern. This may for instance be the case when amoxicillin trihydrate, is mixed with a
5 second pharmaceutically active agent, e.g. clavulanic acid. Therefore, the prior art proposes drying amoxicillin trihydrate to a certain extent.

The water activity, defined as the equilibrium relative humidity divided by 100%, is a method for specifying the extent to which a β -lactam antibiotic has to be dried. The water activity can be measured by bringing a quantity of the sample in a closed chamber
10 having a relatively small volume, measuring the relative humidity as a function of time until the relative humidity has become constant, the latter being the equilibrium relative humidity for that sample. For applications wherein problems associated with water play a role, generally low values for the water activity of the β -lactam antibiotic are specified.

If drying is carried out to an insufficient extent, the problems associated with
15 water remain. If drying is carried out too extensively, the physical properties, such as colour and stability may be impaired. This may for instance be due to the fact that crystal water may be expelled when drying is carried out too extensively.

We surprisingly found a product of amoxicillin trihydrate, having a free water content less than 0.10 wt.%, measured at an equilibrium relative humidity of 30% and at
20 a temperature of 25 °C; and a process for the preparation thereof.

Accordingly a product of amoxicillin trihydrate is provided by the invention giving no or fewer problems with water compared to amoxicillin trihydrate according to the prior art having the same water activity. Moreover, drying may be carried out to a lesser extent, such that properties such as colour and stability are not impaired or impaired to a
25 lesser extent. Due to its lower free water content, the amoxicillin trihydrate, can advantageously be mixed with clavulanic acid or salts thereof, which is known to be very sensitive to moist.

As used herein, the free water content measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C is in particular defined as

30 $((w_{30}-w_{10})/w_{10}) \cdot 100\%$, wherein

w_{30} = sample weight, corresponding to an equilibrium relative humidity of 30% at temperature of 25 °C

w_{10} = sample weight, corresponding to an equilibrium relative humidity of 10% at temperature of 25 °C.

The free water content, including the values for w_{30} and w_{10} are preferably determined using dynamic vapour sorption, for instance using a VTI-SGA 100 vapour sorption analyser. Using this technique, adsorptions terms are preferably measured for a sample having a weight of 200 mg by conditioning the air inside the sample chamber at a relative humidity of 10% for 90 minutes, and subsequently increasing the relative humidity with steps of 10%, maintaining the sample at each value for the relative humidity for 90 minutes and taking the sample weight measured after 90 minutes for a value of the relative humidity as the sample weight corresponding to the equilibrium relative humidity.

The product of amoxicillin trihydrate according to the invention has a free water content less than 0.10 wt.%, measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C. Preferably, the product of amoxicillin trihydrate according to the invention has a free water content less than 0.07 wt.%, more preferably less than 0.05 wt.%, measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C. There is no specific upper limit for the free water content. The free water content may for instance be higher than 0.01 wt.%, measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C.

The product of amoxicillin trihydrate according to the invention may be amoxicillin trihydrate in any suitable form, for instance in the form of crystalline powder or granules or mixtures comprising crystalline powder and granules. In a preferred embodiment, the product according to the invention is crystalline amoxicillin trihydrate powder having a free water content of less than 0.1 wt.%, preferably less than 0.07 wt.%, more preferably less than 0.05 wt.%, measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C.

It will be understood that the product of amoxicillin trihydrate may still contain some impurities. Preferably, the product of amoxicillin trihydrate contains at least 90 wt.%, preferably at least 95 wt.%, more preferably at least 98 wt.% of amoxicillin trihydrate. These weight percentages are given relative to the weight of the product. Preferably, the product of amoxicillin trihydrate according to the invention is free of auxiliaries.

As used herein crystalline amoxicillin trihydrate powder refers in particular to a product consisting mainly of crystals of amoxicillin trihydrate. It will be understood that crystals do not refer to aggregates formed by the build-up of crystals, for instance with the aid of a binding agent like water or starch paste, or mechanical force like roller

compacting or extrusion. Some unintended formation of aggregates may occur during usual handling, for instance during drying. Aggregates can be seen using optical microscopy applied at a magnification of 140x. As used herein a product consisting mainly of crystals of amoxicillin trihydrate refers in particular to a product comprising at least 70 wt.% crystals of amoxicillin trihydrate, preferably at least 80 wt.%, more preferably at least 90 wt.%, more preferably at least 95 wt.%, more preferably at least 98 wt.%. These percentages can be determined using a combination of air jet sieving and optical microscopy. Air Jet sieving is advantageously carried out using an Alpine Air Jet 200LS-N air jet sieve during 1 minute at 1200 Pa for a sample weight of 10 g.

Optical microscopy is advantageously carried out by taking a sample of said fraction of 5 mg, suspending the sample suspending in 4 drops of paraffin oil on a surface on a surface area of 22x40 mm, and using a magnification of 140x.

It will be understood that the amoxicillin trihydrate powder may still contain some impurities. Preferably, the product of amoxicillin trihydrate contains at least 90 wt.%, preferably at least 95 wt.%, more preferably at least 98 wt.% of amoxicillin trihydrate. These weight percentages are given relative to the weight of the crystalline powder. Preferably, the crystalline amoxicillin trihydrate powder according to the invention is free of auxiliaries.

In a preferred embodiment, the product of amoxicillin trihydrate according to the invention, has a water activity of more than 0.05, preferably more than 0.07, preferably more than 0.10, preferably more than 0.15, preferably more than 0.20, preferably more than 0.25, preferably more than 0.30. Increased water contents are advantageous as properties of the amoxicillin trihydrate are not impaired or impaired to a lesser extent, while the amount of free water is still low. There is no specific lower limit for the water activity. In practice, the water activity will generally be less than 0.7, for instance less than 0.6, for instance less than 0.5, although this is not necessary. As used herein these values refer to the water activity at 25 °C.

A preferred method for determining the water activity of a sample is to bring a quantity of the sample in a closed chamber having a relatively small volume, measuring the relative humidity as a function of time until the relative humidity has become constant (for instance after 30 minutes), the latter being the equilibrium relative humidity for that sample. Preferably, a Novasina TH200 Thermoconstanter is used, of which the sample holder has a volume of 12 ml and which is filled with 3 g. of sample.

The d_{10} and d_{50} are known ways for indicating a particle size distribution, d_{50} referring to the value for the particle size such that 50 vol.% of the crystals has a particle size smaller than said value. The d_{50} is also referred to as average volume-based grain size. Likewise, d_{10} refers to the value for the particle size such that 10 vol.% of the crystals has a particle size smaller than said value. A preferred way for determining d_{10} and d_{50} is laser diffraction, preferably using Malvern equipment. A suitable apparatus for determining d_{10} and d_{50} is a Malvern particle sizer 2600 C obtainable from Malvern Instruments Ltd., Malvern UK, using an objective of $f = 300$ mm and a beam length of 14.30 mm. A polydisperse analysis model may advantageously be used.

We found that crystalline amoxicillin trihydrate powder according to the invention preferably has an increased d_{50} . The invention therefore also provides crystalline amoxicillin trihydrate powder, preferably having a d_{50} of higher than $10\text{ }\mu\text{m}$, preferably higher than $20\text{ }\mu\text{m}$, more preferably higher than $30\text{ }\mu\text{m}$, more preferably higher than $35\text{ }\mu\text{m}$, more preferably higher than $40\text{ }\mu\text{m}$. There is no specific upper limit for the d_{50} . The d_{50} of the crystalline powder according to the invention may be less than $150\text{ }\mu\text{m}$, for instance less than $100\text{ }\mu\text{m}$. The crystalline powder according to the invention preferably has increased d_{10} , preferably higher than $3\text{ }\mu\text{m}$, preferably higher than $5\text{ }\mu\text{m}$, more preferably higher than $8\text{ }\mu\text{m}$, more preferably higher than $10\text{ }\mu\text{m}$. There is no specific upper limit for the d_{10} of the crystalline powder according to the invention. The d_{10} of the crystalline powder according to the invention may be less than $50\text{ }\mu\text{m}$.

Crystalline amoxicillin trihydrate powder, may be obtained by preparing a solution comprising dissolved amoxicillin, crystallizing the amoxicillin from said solution to form crystals, separating the crystals from said solution, and drying the separated crystals. As used herein the term crystalline powder includes, but is not limited to, the dried product obtained and/or obtainable by this process.

It was found that preparing crystalline powder having decreased free water content for a given equilibrium relative humidity preferably includes carrying out the

process, in particular the crystallizing, separating and drying, under such conditions that the dried crystals have an increased particle size, in particular increased d_{50} and/or d_{10} .

Preferred crystallization conditions include crystallization conditions such that the amoxicillin trihydrate that crystallizes from the solution has increased particle size. This
5 may for instance be achieved by applying a relatively long residence time, relatively low concentrations of amoxicillin in the aqueous solution or using an aqueous solution having high purity. Further preferred conditions are described hereinafter.

The extent of mechanical impact, for instance during separation and/or drying may influence the particle size. Mechanical impact during separation can for instance be
10 achieved during centrifuging. Mechanical impact during drying can for instance be achieved by using a contact dryer, for instance a Vrieco-Nauta contact dryer, or a flash dryer. Mechanical impact can also be achieved by applying pneumatic transport, for instance pneumatic transport of the amoxicillin trihydrate from the separation step to the drying step. A too high extent of mechanical impact is may result in an undesirable
15 decrease of the particle size. Using this insight provided by the invention and by varying the mechanical forces the skilled man is able to find out the conditions how an undesired reduction of particle size can be avoided.

In view of the above, the invention also provides a process for preparing crystalline amoxicillin trihydrate powder, said process comprising: crystallizing amoxicillin
20 trihydrate from a solution; separating the crystals from said solution; drying the separated crystals; wherein the process, preferably the crystallizing, separating and/or drying, is carried out under such conditions that the resulting crystalline powder has a d_{50} of higher than 10 μm , preferably higher than 20 μm , more preferably higher than 30 μm , in particular higher than 35 μm , more preferably higher than 40 μm . There is no
25 specific upper limit for the d_{50} . The process, preferably the crystallizing, separating and/or drying, may for instance be carried out under such conditions the d_{50} of the resulting crystalline powder is less than 150 μm , for instance less than 100 μm . Preferably, the process, preferably the crystallizing, separating and/or drying, is carried out under such conditions that the dried crystals have a d_{10} of higher than 3 μm ,
30 preferably higher than 5 μm , more preferably higher than 8 μm , more preferably higher than 10 μm . There is no specific upper limit for d_{10} . The process, preferably the crystallizing, separating and/or drying, may be carried out under such conditions that d_{10} of the resulting crystalline powder is less than 50 μm .

We found that crystals of amoxicillin trihydrate having decreased free water content may preferably be obtained by applying preferred process conditions described hereinafter.

Preferably, a process for preparing crystalline amoxicillin trihydrate powder according to the invention comprises preparing amoxicillin by reacting 6-amino-
5 penicillanic acid or a salt thereof, with para-hydroxyphenyl glycine in activated form in the presence of an enzyme immobilized on a carrier; forming an aqueous solution containing the amoxicillin, said aqueous solution containing hydrochloric acid; and crystallizing the amoxicillin trihydrate from said aqueous solution.

10 Preferably, the solution from which the amoxicillin trihydrate is crystallized is an aqueous solution. Any suitable aqueous solution may be used. Suitable aqueous solutions include solutions wherein the weight ratio water: organic solvent is between 100 : 0 and 70 : 30, preferably between 100 : 0 and 80 : 20, preferably between 100 : 0 and 90 : 10, preferably between 100 : 0 and 95 : 5, preferably between 100 : 0 and
15 99 : 1.

Preferably, the solution from which the amoxicillin trihydrate is crystallized contains less than 200 weight parts of protein per 1.000.000 weight parts of amoxicillin (total concentration of amoxicillin, whether or not in dissolved form), preferably less 100 weight parts of protein, more preferably less than 50 weight parts of protein, more
20 preferably less than 35 weight parts of protein.

Preferably, the solution from which the amoxicillin trihydrate is crystallized is an aqueous solution having an amoxicillin concentration (total concentration of amoxicillin, whether or not in dissolved form) of less than 0.6 mol/l, preferably less than 0.5 mol/l, more preferably less than 0.4 mol/l, more preferably less than 0.3 mol/l.

25 The aqueous solution from which the amoxicillin trihydrate is crystallized, is preferably a solution containing hydrochloric acid or chloride. The aqueous solution from which the amoxicillin trihydrate is crystallized preferably contains between 0.9 and 5 mol of hydrochloric acid or chloride per mol amoxicillin (total concentration of amoxicillin, whether or not in dissolved form), preferably between 0.9 and 3 mol hydrochloric acid or
30 chloride per mol amoxicillin, more preferably between 0.9 and 1.5 mol hydrochloric acid or chloride per mol amoxicillin. The aqueous solution from which the amoxicillin is crystallized preferably contains more than 1.0 mol of hydrochloric acid or chloride per mol of amoxicillin.

Preferably, the amoxicillin trihydrate is crystallized from an aqueous solution at a pH of between 2 and 7, preferably between 3 and 6. Preferably, the process comprises crystallizing amoxicillin trihydrate from the aqueous solution in a first step preferably at a pH of between 2 and 5, preferably between 3 and 4, and in a second step at a pH higher than in the first step, preferably between 4 and 7, preferably between 4.5 and 6.

Preferably, amoxicillin trihydrate is crystallized from the aqueous solution at a temperature of between 5 °C and 40 °C, preferably between 10 and 30 °C, more preferably between 15 and 25 °C.

The solution from which the amoxicillin trihydrate is crystallized may be prepared in any suitable way. The aqueous solution containing dissolved amoxicillin may be prepared by dissolving amoxicillin trihydrate. It is possible to add amoxicillin trihydrate to a solution, and to effect dissolution of the added amoxicillin trihydrate. It is also possible to prepare an aqueous suspension by forming crystals of amoxicillin trihydrate in situ in a solution, and effecting dissolution of the crystals of amoxicillin trihydrate in said suspension. In a process for the preparation of amoxicillin, the process preferably comprises preparing an aqueous solution containing dissolved amoxicillin, said aqueous solution having an amoxicillin concentration of less than 0.6 mol/l, preferably less than 0.5 mol/l, more preferably less than 0.4 mol/l, more preferably less than 0.3 mol/l. The process preferably comprises preparing an aqueous solution containing dissolved amoxicillin, said aqueous solution having a pH of between 0 and 1.5, preferably between 0.5 and 1.2. Dissolving amoxicillin, may be carried out in any suitable way, for instance by adding an acid, preferably by adding hydrochloric acid to an aqueous suspension containing crystals of amoxicillin trihydrate. An acid, preferably hydrochloric acid, may be added in an amount of between 0.9 and 5 mol of hydrochloric acid per mol amoxicillin, preferably between 0.9 and 3 mol hydrochloric acid per mol amoxicillin, more preferably between 0.9 and 1.5 mol hydrochloric acid per mol amoxicillin. Preferably more than 1.0 mol of hydrochloric acid is added per mol of amoxicillin. In a preferred embodiment, the process comprises keeping the (aqueous) solution or (aqueous) suspension at a pH of less than 1.5, preferably less than 1.2, during a period of less than 60 minutes, preferably less than 30 minutes, more preferably less than 15 minutes, more preferably less than 10 minutes, more preferably less than 8 minutes, as this may improve the purity of the amoxicillin. Preferably, the process comprises mixing the aqueous solution or aqueous suspension with the acid using a fast mixer, for instance a static mixer. This can reduce the time during which the aqueous solution or suspension

is kept low pH. Mixing of the acid with the aqueous suspension may be carried out at any suitable temperature, for instance higher than -5°C , for instance higher than 5°C , for instance higher than 10°C , for instance higher than 15°C , for instance less than 50°C , for instance less than 40°C . Preferably, the process comprises filtering the solution prior to said crystallizing. Preferably, the process comprises filtering the aqueous solution containing dissolved amoxicillin, said aqueous solution preferably having a pH of between 0 and 1.5, preferably between 0.5 and 1.2. The solution may be passed through any suitable filter. Preferably, a filter is used having a pore size of less than $40\text{ }\mu\text{m}$, preferably less than $20\text{ }\mu\text{m}$, preferably less than $10\text{ }\mu\text{m}$, and more preferably less than $5\text{ }\mu\text{m}$.

Amoxicillin trihydrate may advantageously be crystallized from said aqueous solution by increasing the pH, for instance by adding a base, for instance NaOH.

The crystallization may be carried out batch wise or continuously. When the process is carried out batch wise, addition of seed crystals to the aqueous solution is preferred. Preferably, the crystallization is carried out continuously.

Amoxicillin is preferably prepared by reacting 6-amino-penicilanic acid or derivatives thereof, for instance a salt of 6-amino-penicilanic acid, with an acylating agent selected from para-hydroxyphenyl glycine in activated form in the presence of an enzyme in an aqueous reaction medium. The para-hydroxyphenyl glycine in activated form is preferably an ester or amide of para-hydroxyphenylglycine. Suitable esters include for instance 1 to 4 alkyl esters, for example methyl ester, ethyl ester, n-propyl or isopropyl esters. Glycol esters, for instance an ethylene glycol ester, may also be used. An amide that is unsubstituted in the $-\text{CONH}_2$ group may be used.

The enzyme may be any enzyme having hydrolytic activity (hydrolase). The enzyme may for instance be an acylase, *inter alia* Penicillin G acylase, amidase or esterase. Enzymes may be isolated from various naturally occurring microorganisms, for example fungi and bacteria. Organisms that have been found to produce penicillin acylase are, for example, *Acetobacter*, *Aeromonas*, *Alcaligenes*, *Aphanocladium*, *Bacillus* sp., *Cephalosporium*, *Escherichia*, *Flavobacterium*, *Kluyvera*, *Mycoplana*, *Protaminobacter*, *Pseudomonas* or *Xanthomonas* species.

Processes for the preparation of amoxicillin in the presence of an enzyme have been described in WO-A-9201061, WO-A-9417800, WO-A-9704086, WO-A-9820120, EP-A-771357, the contents of which are incorporated by reference.

The reaction may be carried out at any suitable pH, preferably at a pH of between 5 and 9, preferably between 5.5 and 8, more preferably between 6 and 7.5. The reaction may be carried out at any suitable temperature, for instance carried out at a temperature of between 0 and 40 °C, preferably between 0 and 30 °C, more preferably
5 between 0 and 15 °C.

The amoxicillin formed may be crystallized under the conditions at which the reaction is carried out. Crystallization of amoxicillin may for instance be effected at a pH of between 5 and 8, preferably between 5.5 and 7.5.

Preferably, the enzyme is an enzyme immobilized on a carrier. Any suitable
10 carrier may be used. Preferably, the carrier comprises a gelling agent and a polymer containing free amino groups. Preferably, the polymer is selected from alginate amine, chitosan, pectin, or polyethylene imine. Preferably, the gelling agent is gelatin. This carrier and the preparation thereof are described in EP-A-222 462 and WO-A-9704086. Prior to immobilization, the isolated enzyme is preferably purified using ion exchange
15 chromatography.

Preferably, the enzyme is an enzyme immobilized on a carrier, and the process preferably comprises separating a product comprising the amoxicillin formed from the immobilized enzyme. Said separating of the product from the immobilized enzyme may be carried out using any suitable method, for instance by using gravity or a screen that
20 is not permeable to the major part of the immobilized enzyme. Preferably, the product separated from the immobilized enzyme contains less than 200 weight parts of protein per 1.000.000 weight parts of the amoxicillin, preferably less 100 weight parts of protein, more preferably less than 50 weight parts of protein, more preferably less than 35 weight parts of protein per 1.000.000 weight parts of the amoxicillin. This is preferably
25 achieved by applying an enzyme sufficiently immobilized on a carrier to avoid separation of small amounts of protein from amoxicillin trihydrate. This embodiment has the advantage that the final amoxicillin trihydrate obtained contains less than 200 weight parts of protein per 1.000.000 weight parts of the amoxicillin, preferably less 100 weight parts of protein, more preferably less than 50 weight parts of protein, more preferably
30 less than 35 weight parts of protein per 1.000.000 weight parts of amoxicillin. The product separated from the immobilized enzyme may be an aqueous solution containing amoxicillin in dissolved form. The product separated from the immobilized enzyme may also be a wet cake. The separated product is preferably an aqueous suspension comprising amoxicillin trihydrate crystals. Preferably, the process comprises dissolving

said amoxicillin trihydrate crystals to form an aqueous solution containing dissolved amoxicillin.

The invention also relates to crystalline amoxicillin trihydrate powder obtainable by the process according to the invention.

5 The product according to the invention can advantageously be used for the preparation of a pharmaceutical composition.

The product of amoxicillin trihydrate according to the invention can advantageously be mixed with pharmaceutically acceptable auxiliaries and/or with a second pharmaceutically active agent. The product of amoxicillin trihydrate, according to
10 the invention can for instance be mixed with between 0 and 50 wt.%, preferably between 0 and 40 wt.%, preferably between 0 and 30 wt.%, more preferably between 0 and 20 wt.%, preferably more than 1 wt.% of auxiliaries, relative to the sum weight of the crystalline powder and the auxiliaries. The product of amoxicillin trihydrate can for instance be mixed with clavulanic acid in salt form, preferably as a potassium salt, the
15 weight ratio amoxicillin : clavulanic acid preferably being between 1:1 and 15:1, preferably between 2:1 and 10:1, preferably between 4:1 and 8:1. These weight ratios are calculated for the anhydrous amoxicillin and clavulanate in acid form. Therefore, the invention also relates to a mixture obtainable by a process comprising mixing the product of amoxicillin trihydrate according to the invention with auxiliaries and/or a
20 second pharmaceutically active agent. The invention also provides a mixture comprising (i) the product of amoxicillin trihydrate powder according to the invention and (ii) a second pharmaceutically active agent, with or without auxiliaries.

As a second pharmaceutically active agent, clavulanic acid in the form of a salt, preferably clavulanic acid in the form of a potassium salt is preferably used.

25 As auxiliaries may for instance be used fillers, dry binders, disintegrants, wetting agents, wet binders, lubricants, flow agents and the like. Examples of auxiliaries are lactose, starches, bentonite, calcium carbonate, mannitol, microcrystalline cellulose, polysorbate, sodium lauryl sulphate, carboxymethylcellulose Na, Sodium alginate, magnesium stearate, silicon dioxid, talc.

30 In an embodiment, the mixture contains between 0 and 50 wt.%, preferably between 0 and 40 wt.%, preferably between 0 and 30 wt.%, more preferably between 0 and 20 wt.%, preferably more than 1 wt.% of auxiliaries. These weight percentages are given relative to the sum weight of the amoxicillin trihydrate and the auxiliaries.

Preferably the weight ratio amoxicillin : clavulanic acid is between 1:1 and 15:1, preferably between 2:1 and 10:1, preferably between 4:1 and 8:1. These weight ratios are calculated for the anhydrous amoxicillin and clavulanate in acid form.

The product or the mixture according to the invention can advantageously be
5 used for filling a capsule for pharmaceutical use, for instance a gelatine capsule.
Therefore, the invention also relates to a capsule containing the product according to the invention or a capsule containing the mixture according to the invention. The product according to the invention or the mixture according to the invention may be fed into a capsule in any suitable way. The invention also relates to the use of the product
10 according to the invention or the mixture according to the invention for filling a capsule.

The invention also provides a process comprising compressing the product according to the invention or compressing the mixture according to the invention to produce compressed products. The compressed products may for instance be granules or tablets. The invention also relates to granules or tablets comprising the product
15 according to the invention in compressed form or comprising the mixture according to the invention in compressed form.

The invention also relates to a process for preparing granules, comprising feeding the crystalline powder according to the invention or the mixture according to the invention, optionally in combination with auxiliaries and/or a second pharmaceutically
20 active agent, to a roller compactor to produce compacts; and milling the compacts to produce granules. The granules produced may advantageously be sieved to obtain a desired particle size distribution. The invention also relates to granules obtainable by this process.

The invention also relates to a process for preparing granules, comprising mixing
25 the crystals according to the invention or the mixture according to the invention with a binding agent, the binding agent for instance being dissolved in a moistening liquid; compacting the crystals whilst moist or dry; granulating the compacts obtained through a sieve. The invention also relates to granules obtainable by this process.

The invention also relates to a process comprising forming a paste from the
30 crystalline powder according to the invention or from the mixture according to the invention; kneading the paste at a temperature of 10 °C to 80 °C; extruding the paste in a double-screwed extruder, and, if desired, drying the granules obtained. The invention also relates to granules obtainable by this process.

The invention also relates to a process comprising compressing the granules according to the invention, optionally in mixture with auxiliaries and/or a pharmaceutically active agent to prepare tablets. The invention also relates to tablets obtainable by this process.

5 We also found that the physical properties of crystalline amoxicillin trihydrate powder can be improved as regards flowability.

In a preferred embodiment, the amoxicillin trihydrate powder according to the invention has a bulk density higher than 0.45 g/ml. The crystalline powder according to this aspect of the invention has improved flow properties, without having to
10 be subjected to processes such as granulation, compactation, agglomeration or aggregation, has good colour properties, good stability and a high dissolution rate. If it would still be desired to subject the crystalline powder to processes such as granulation, compactation, agglomeration or aggregation, and the like, applying these processes is facilitated due to the improved flow properties of the crystalline powder according to this
15 aspect of the invention. Moreover, an increased quantity of crystalline powder can be fed into a capsule of a given size. The bulk density is preferably determined according to USP 24, method I, (page 1913). Preferably, of the crystalline powder has a bulk density higher than 0.46 g/ml, preferably higher than 0.5 g/ml, more preferably higher than 0.55 g/ml. This further improves the flow properties. Moreover, an increased bulk density is
20 advantageous as crystalline powder may be fed into a certain volume, for instance a capsule. There is no specific upper limit for the bulk density. The bulk density may be less than 0.8 g/ml, for instance less than 0.7 g/ml.

In a preferred embodiment, the crystalline powder according to the invention has a tapped density higher than 0.6 g/ml, preferably higher than 0.7, more preferably higher
25 than 0.8 g/ml. An increased tapped density improves the flow properties. Moreover, an increased tapped density is advantageous as more products may be fed into a certain volume, for instance a capsule. There is no specific upper limit for the tapped density. The tapped density may be less than 1.2 g/ml, for instance less than 1.1 g/ml, for instance less than 1.0 g/ml. The tapped density is preferably determined according to
30 USP 24, method II, (page 1914).

In a preferred embodiment, the crystalline powder according to the invention has a bulk density and tapped density such that the ratio d_t/d_b is less than 1.7, preferably less than 1.6, preferably less than 1.5, preferably less than 1.45, wherein d_t = tapped density and d_b = bulk density. This results in improved flow ability. There is no specific

lower limit for the ratio d_t/d_b . The ratio d_t/d_b may be higher than 1.05, for instance higher than 1.1.

In a preferred embodiment, the crystalline powder according to the invention has a bulk density and tapped density such that the ratio d_t/d_b is less than 1.7, preferably less than 1.6, preferably less than 1.5, preferably less than 1.45. This crystalline powder has improved flow properties compared to the known powder. There is no specific upper limit for the ratio d_t/d_b . The ratio d_t/d_b may be higher than 1.05, for instance higher than 1.1.

In a preferred embodiment, the crystalline powder according to the invention has a bulk density and tapped density such that the compressibility index as defined by $((d_t - d_b)/d_t) \cdot 100\%$ is less than 40%, preferably less than 35%, more preferably less than 30%. This results in improved flow ability. There is no specific lower limit for the compressibility index. The compressibility index may for instance be higher than 10%.

We found that crystalline powder having improved flowability, bulk density and/or tapped density preferably have an increased d_{50} .

It was surprisingly found that crystalline powder having improved flow properties, in particular the high bulk density and/or high tapped density, can be obtained by selecting the crystallizing, separation and/or drying conditions.

It was found that preparing crystalline powder having improved flow properties, in particular high bulk density and/or high tapped density, preferably includes carrying out the process, in particular the crystallizing, separating and drying, under such conditions that the dried crystals have an increased particle size, in particular increased d_{50} and/or d_{10} .

It was further found that, in particular for crystals having an increased size, the extent of mechanical impact, for instance during crystallization, separation and/or drying influences the bulk density and tapped density. If the crystals are subjected to mechanical forces, for instance during drying and/or separating or transport of the crystals, the bulk density and tapped density are surprisingly found to increase compared to the situation where there is no mechanical impact. However, if the mechanical forces are too high, the bulk density and tapped density are found to decrease. Mechanical impact during separating can for instance be achieved during centrifuging. Mechanical impact during drying can for instance be achieved by using a contact dryer, for instance a Vrieco-Nauta contact dryer, or a flash dryer. Mechanical impact can also be achieved by applying pneumatic transport, for instance pneumatic

transport of the amoxicillin trihydrate from the separation step to the drying step. Without wishing to be bound by any scientific theory, it is believed that a limited extent of mechanical impact has the effect that relatively large crystals having the shape of needles are broken, thereby resulting in an increase of the bulk density and/or tapped density. However, a too high extent of mechanical forces is believed to result in the production of crystals that are too fine, thereby reducing the bulk density and/or tapped density. Using this insight provided by the invention and by varying the mechanical forces the skilled man is able to find out the conditions where the optimal bulk density and/or tapped density are achieved.

The invention also provides a process comprising sieving the crystalline powder according to the invention. This allows the physical properties of the crystalline powder to be improved even further. Preferably, air jet sieving is applied.

The invention will be further elucidated by means of the following examples, without however being limited thereto.

Examples and Comparative experiment:

Preparation of immobilized enzyme

Escherichia coli penicillin acylase was isolated as described in WO-A-9212782, purified using ion exchange chromatography, and immobilized as described in EP-A-222462 and WO-A-9704086.

As definition of penicillin G acylase activity the following is used: one unit (U) corresponds to the amount of enzyme that hydrolyses per minute 1 μ mole penicillin G under standard conditions (100 g.l⁻¹ Penicillin G potassium salt, 0.05 M potassium phosphate buffer, pH value 8.0, 28 °C).

Production of amoxicillin

162.2 g of 6-APA (6-amino-penicillanic acid) and 184.8 g of HPGM (D(-)-p-hydroxyphenylglycine methyl ester) were suspended in 450 ml of water. The suspension was cooled to a temperature of 10°C. To this reaction mixture 32850 Units of immobilized penicillin acylase were added, and water was added to a final volume of 1500 ml. The mixture was stirred for 6 hours. During the reaction the pH increased to

6.9, and at the end of the reaction the pH had decreased to 6.2. To this mixture 750 ml of water was added, and the suspension was filtrated over a sieve (with a mesh of 100 micrometer) in 2 hours to separate off the immobilized enzyme. The suspension obtained containing the amoxicillin trihydrate crystals was cooled to 0°C. The
5 suspension contained less than 50 ppm of protein relative to amoxicillin trihydrate (less than 50 weight parts of protein per 1.000.000 weight parts of amoxicillin trihydrate).

An aqueous suspension containing amoxicillin in water (100 g amoxicillin trihydrate per liter of suspension) as obtained above was mixed with a 32 wt.% HCl solution (at a
10 temperature of 25 °C) using a static mixer such as to obtain a solution having a pH of 1. The residence time in the static mixer was 1.5 minutes. The acidic solution obtained is pumped through two filters, the first filter having pores of 40 μ m, the second filter having pores of 4.5 μ m. The residence time in the filters was about 3 minutes. The acidic filtered solution and is fed to a first stirred tank in which a pH of 3.7 is maintained by
15 addition of a 8 M NaOH solution. The temperature in the first tank is between 17 and 23 °C. The residence time in the first tank is 45 minutes. The contents of the first tank are fed to a second stirred tank in which a pH of 5.0 is maintained by addition of a 8 M NaOH solution. The temperature in the second stirred tank is between 17 and 23 °C. The residence time in the second stirred tank is 15 minutes. The contents of tank 2 fed
20 to a third stirred tank in which the a temperature of 1 to 5 °C is maintained, the residence time in the third tank being more than 4 hours. The contents of the third stirred tank are fed to an inverted filter centrifuge such as to isolate the amoxicillin crystals, resulting in a wet cake containing 86 wt.% solid material. The wet cake was washed with water, pneumatically transported to a conical vacuum contact drier (Vrieco-
25 Nauta), in which it was dried at a temperature of 30 to 40 °C and a pressure of 30 mbar during 7 hours.

Measurement of particle size distribution.

The particle size distribution (including d_{10} and d_{50}) was determined using a
30 Malvern particle sizer 2600 C with objective $f = 300$ mm, malvern sample measurement unit PS1 and a Malvern dry powder feeder PS 64. The beam length was 14.30 mm. A polydisperse analysis model was used.

Measurement of adsorption isotherms

Adsorption isotherms were determined using dynamic vapour sorption, using a VTI-SGA 100 vapour sorption analyser. Samples were used having a weight of 200 mg. Air inside the sample chamber was conditioned at a relative humidity of 10% for 90 minutes. Subsequently, the relative humidity was increased with steps of 10%, while maintaining the sample was held at each value for the relative humidity for 90 minutes. The weight of the sample after said 90 minutes was taken as the sample weight corresponding to the equilibrium relative humidity. The temperature was 25 °C.

Example I

A batch of amoxicillin trihydrate crystalline powder was prepared using the process as described above. The adsorption isotherm, as well as the particle size distribution was determined. The adsorption isotherm is shown in figure I. The d_{50} and d_{10} are indicated in the table.

The free water content, measured at an equilibrium relative humidity of 30% at 25 °C was 0.05 wt.%.

Comparative experiment A

In a chemical process for the preparation of amoxicillin, a solution containing amoxicillin in dilute HCl and isopropanol was obtained. This solution was fed to a stirred tank. The pH was kept at 3.7 at a temperature of 20 °C. Subsequently the pH was raised to 5.0 by adding NaOH. The resulting mixture is maintained in a vessel at 1 to 5 °C during 3 to 12 hours. The amoxicillin was separated using a centrifuge, and dried using a fluid bed dryer.

The adsorption isotherm, as well as the particle size distribution was determined. The adsorption isotherm is shown in figure I. The d_{50} and d_{10} are indicated in the table.

The free water content, measured at an equilibrium relative humidity of 30% at 25 °C was 0.11 wt.%.

	Bulk dens. (g/ml)	Tap. dens. (g/ml)	d ₅₀ (μm)	d ₁₀ (μm)	Free water content at ERH = 30% (in wt.%)
Ex. I	0.51	0.73	43.1	11.4	0.05 wt. %
Comp. A			8.3	2.7	0.11 wt. %

Comparison of example I and comparative experiment A shows that the amoxicillin powder of example I contains less free water than the amoxicillin powder of comparative experiment A.

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Example II

Amoxicillin trihydrate powder obtained by the process of example I is dried to a water activity of 0.15. The powder is mixed with potassium clavulanate in a weight ratio 4 to 1 (calculated for the amoxicillin anhydrate and clavulanic acid). The mixture is stable.

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Example III

Amoxicillin powder obtained by the process of example I is dried to a water activity of 0.2. The powder is mixed with potassium clavulanate in a ratio 4 to 1. The mixture is stable.

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Example IV

Amoxicillin powder obtained by the process of example I is dried to a water activity of 0.15. The powder is mixed with potassium clavulanate in a ratio 4 to 1. The mixture is stable.

Example V

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Example I was repeated with the difference that drying was not carried out using the a conical vacuum contact drier (Vrieco Nauta), but using a drier wherein the material was not mechanically impacted (Ventilation stove). Drying was carried out at a temperature of 35 °C during 16 hours. The d₅₀ and d₁₀ are 66.3 μm and 17.4 μm respectively. The bulk density and the tapped density are 0.25 g/ml and 0.47 g/ml respectively.

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Example VI

Another batch of amoxicillin trihydrate powder was prepared according to example I (d₅₀ and d₁₀ are 61 μm and 19 μm respectively, bulk density and the tapped density are 0.58 g/ml and 0.79 g/ml respectively). This batch was subjected to air jet sieving (200 LS-N air

jet sieve manufactured by Hosakawa Alpine). Sieving was carried out using a 75 μm screen during 10 minutes. A few agglomerates formed during said sieving were removed from the overheads fraction (not passed through the sieve) using a vibrating sieve (425 μm), after which tapped density, bulk density, d_{50} , d_{10} of the resulting crystals of the overheads fraction was determined. The d_{50} and d_{10} are 86 μm and 36 μm respectively. The bulk density and the tapped density are 0.59 g/ml and 0.74 g/ml respectively.

CLAIMS

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1. Product of amoxicillin trihydrate, having a free water content of less than 0.1 wt.%, measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C.

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2. Product according to claim 1, wherein said product has a free water content of less than 0.07 wt.%, more preferably less than 0.05 wt.%, measured at an equilibrium relative humidity of 30% and at a temperature of 25 °C.

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3. Product according to claim 1, having a water activity of more than 0.05, preferably more than 0.07, preferably more than 0.10, preferably more than 0.15, preferably more than 0.20, preferably more than 0.25, preferably more than 0.30, measured at a temperature of 25 °C.

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4. Product according to any one of claims 1 to 3, wherein said product is crystalline amoxicillin trihydrate powder.

5. Crystalline powder according to claim 4, having a d_{50} higher than 10 μm , preferably higher than 20 μm , more preferably higher than 30 μm , more preferably higher than 35 μm , more preferably higher than 40 μm .

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6. Crystalline powder according to claim 4 or claim 5, having a d_{10} of higher than 3 μm , preferably higher than 5 μm , more preferably higher than 8 μm , more preferably higher than 10 μm .

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7. Crystalline powder according to any one of claims 4 to 6, having a bulk density higher than 0.45 g/ml, preferably higher than 0.5 g/ml, more preferably higher than 0.55 g/ml.

8. Crystalline powder according to any one of claims 4 to 7 having a tapped density higher than 0.6 g/ml, preferably higher than 0.7, more preferably higher than 0.8 g/ml.

9. Process comprising mixing the product according to any one of claims 1 to 8 with a second pharmaceutically active agent, and/or auxiliaries.

10. Mixture comprising
(i) the product according to any one of claims 1 to 8; and
(ii) a second pharmaceutically active agent, and/or auxiliaries.

11. Process according to claim 9 or mixture according to claim 10, wherein said second pharmaceutically active agent is clavulanic acid in the form of a salt, preferably clavulanic acid in the form of a potassium salt.

12. Process comprising compressing the product according to any one of claims 1 to 8 or the mixture according to claim 10 to produce compressed products.

13. Process according to claim 12, wherein said compressed products are granules or tablets.

14. Use of the product according to any one of claims 1 to 8 for the preparation of a pharmaceutical composition.

15. Use of the product according to any one of claims 1 to 8 or the mixture according to claim 10 for the preparation of a tablet or for filling a capsule.

16. Process for preparing amoxicillin trihydrate, said process comprising:
preparing amoxicillin by reacting 6-amino-penicillanic acid or a salt thereof, with para-hydroxyphenyl glycine in activated form in the presence of an enzyme immobilized on a carrier;
forming an aqueous solution containing the amoxicillin, said aqueous solution containing hydrochloric acid; and
crystallizing amoxicillin trihydrate from said aqueous solution.

17. Process according to claim 16, wherein the aqueous solution from which the amoxicillin is crystallized has an amoxicillin concentration of less than 0.6 mol/l, preferably less than 0.5 mol/l, more preferably less than 0.4 mol/l, more preferably less than 0.3 mol/l.

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18. Process according to claim 16 or claim 17, wherein the solution from which the amoxicillin is crystallized contains less than 200 weight parts of protein per 1.000.000 weight parts of the amoxicillin in said solution, preferably less 100 weight parts of protein, more preferably less than 50 weight parts of protein, more preferably less than 35 weight parts of protein.

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19. Process according to any one of claims 16 to 18, wherein the process comprises separating the crystals from said aqueous solution; and drying the separated crystals, resulting in crystalline powder having a d_{50} higher than 10 μm , preferably higher than 20 μm , more preferably higher than 30 μm , more preferably higher than 35 μm , more preferably higher than 40 μm .

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20. Process according to any one of claims 16 to 19, resulting in crystalline powder having a d_{10} higher than 3 μm , preferably higher than 5 μm , more preferably higher than 8 μm , more preferably higher than 10 μm .

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