A NOVEL PHARMACEUTICAL FORMULATION OF ALPHA KETOANALOGUE OF AMINO ACIDS AND ADDITIONAL AMINO ACIDS FOR TREATMENT OF CHRONIC KIDNEY DISEASE AND METHOD OF PREPARATION THEREOF

A novel pharmaceutical formulation of alpha ketoanologue of amino acids and additional amino acids for treatment of chronic kidney disease and method of preparation thereof. The present invention relates to a pharmaceutical formulation having increased in-vitro dissolution, and reduced and controlled degradation under moisture and heat conditions comprising alpha ketoanologue of amino acids and additional amino acids as active ingredients, and disintegrant, filler and binder, and coated with moisture barrier coating composition, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone, and moisture barrier coating composition comprises film former, plasticizer and solubilizing agent, and stabilizer and suspension agent, wherein film former is polyvinyl alcohol, plasticizer and solubilizing agent is soya lecithin, and stabilizer and suspension agent is xanthan gum. In one embodiment, the present invention also relates to a process for preparation of said pharmaceutical formulation.
Title of the Invention:-


5 Field of the Invention:-

The present invention relates to a novel pharmaceutical formulation of alpha ketoanalogue of amino acids and additional amino acids for treatment of chronic kidney disease and method of its preparation. In particular, the present invention relates to a novel pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids which have been surprisingly and unexpectedly found to be effective in improving controlled degradation and enhancing *in-vitro* dissolution of the formulation, particularly to overcome problem of decreased *in-vitro* dissolution of the tablet form of the formulation, more particularly on storage thereof.

In one particular embodiment, the present invention relates to formulation comprising alpha ketoanalogue of amino acids and additional amino acids which have been surprisingly and unexpectedly found to be effective against moisture and heat degradation, and effective to enhance *in-vitro* dissolution of the formulation meaning thereby enhancing availability of the active ingredient of the formulation for efficient treatment of chronic kidney disease.

In another embodiment, the present invention relates to method of preparation of formulation comprising alpha ketoanalogue of amino acids and additional amino acids having controlled degradation and enhanced *in-vitro* dissolution.

Background of the Invention:-

The renal failure or kidney failure (formerly referred as renal insufficiency or chronic renal insufficiency) describes a medical condition in which the kidneys fail to adequately filter toxins and waste products from the blood, which may also be referred to as malfunction of kidney. The problems, frequently encountered in kidney malfunction include abnormal levels of fluid, potassium, calcium, phosphate, hematuria (blood in the urine) in the body, and deranged acid levels, and in the longer term it may lead to anemia.

The long-term kidney problems may have significant repercussions on other diseases, such as cardiovascular disease, and a number of other diseases or health problems may cause renal failure to occur.

The renal failure is typically of two forms - a) acute kidney injury and b) chronic kidney disease, which may be determined by the trend in the serum creatinine level. Other
factors which may help differentiate acute kidney injury from chronic kidney disease include anemia and the kidney size on ultrasound. Chronic kidney disease generally leads to anemia and small kidney size.

The Acute Kidney Injury (AKI) also referred to as acute renal failure (ARF), is a rapidly progressive loss of renal function, generally characterized by oliguria (decreased urine production, quantified as less than 400 mL/day in adults, less than 0.5 mL/kg/h in children or less than 1 mL/kg/h in infants); body water and body fluids disturbances; and electrolyte derangement. AKI can result from a variety of causes, generally classified as prerenal, intrinsic, and postrenal. An underlying cause must be identified and treated to arrest the progress, and dialysis may be necessary to bridge the time gap required for treating these fundamental causes.

The Chronic Kidney Disease (CKD) can develop slowly. Initially it shows few symptoms. The CKD can be the long-term consequence of irreversible acute disease or part of a disease progression.

In case of Acute-on-Chronic Renal Failure (AoCRF), the acute kidney injuries can be present on top of chronic kidney disease. The acute part of AoCRF may be reversible, and the goal of treatment, as with AKI, is to return the patient to baseline renal function, typically measured by serum creatinine. Like AKI, the AoCRF can be difficult to distinguish from chronic kidney disease if the patient has not been monitored by a physician and no baseline (i.e., past) blood work is available for comparison.

Biochemically, the renal failure is typically detected by an elevated serum creatinine level. In the science of physiology, the renal failure is described as a decrease in the glomerular filtration rate (GFR).

All individuals with a GFR less than 60 mL/min/1.73m² for three months are classified as having chronic kidney disease, irrespective of the presence or absence of kidney damage. The rationale for including these individuals in category of chronic kidney disease is that reduction in kidney function to this level or lower represents loss of half or more of the adult level of normal kidney function, which may be associated with a number of complications. The GFR may be sustained at normal or increased levels despite substantial kidney damage and that patients with kidney damage are at increased risk of the two major outcomes of chronic kidney disease, that is, loss of kidney function and development of cardiovascular disease. The loss of protein in the urine is regarded as an independent marker for worsening of renal function and cardiovascular disease. The British guidelines append the letter "P" to the stage of chronic kidney disease if there is
significant protein loss. The level of GFR is generally employed to identify the stage of renal failure.

Stage-I

The normal or relatively high GFR, that is GFR being equal to or more that 90 mL/min/1.73m² indicates slightly diminished function of kidney. This level of kidney damage is defined as pathologic abnormalities or markers of damage, including abnormalities in blood or urine test or imaging studies.

Stage-II

The mild reduction in GFR, that is GFR being between 60 to 89 mL/min/1.73m² indicates kidney damage, which is defined as pathologic abnormalities or markers of damage, including abnormalities in blood or urine test or imaging studies.

Stage-III

The moderate reduction in GFR, that is, GFR being between 30 to 59 mL/min/1.73m². The British guidelines distinguish between stage 3A having GFR between 45 to 59, and stage 3B having GFR between 30 to 44 for the purposes of screening and referral.

Stage-IV

The severe reduction in GFR, that is, GFR being between 15 to 29 mL/min/1.73m². This stage indicates requirement for preparation for renal replacement therapy.

Stage-V

The further severe reduction in GFR, that is, GFR being less than 15 mL/min/1.73m². This stage indicates requirement for permanent renal replacement therapy (RRT).

The formulation comprising alpha ketoanalogue has been used for treatment of renal failure, and for dialysis of patients as a supplement to a protein unrestricted diet in order to compensate amino acid losses in to dialysis. The additional phosphate-binding effects of ketoanalogue associated with reductions in the serum phosphate and parathyroid hormone levels have been shown in dialysis patient.

The alpha ketoanalogue formulation as known in the art under name Ketosteril is combination of alpha ketoanalogue of amino acids and some other amino acids but such combination has been found to give very poor dissolution, and hence, results into inefficient quality for the renal treatment. Further, this known combination which is available in form of film coated tablet has been found to have poor disintegration in the
aqueous medium even after taking for more than one hour, and hence, poor in-vitro dissolution. The inventors have observed that the disintegration time of the formulation known as Ketosteril is more than one hour in water as well as gastric juice at a pH of about 1.2. Further, another known combination of alfa ketoanalogue amino acids [Ketolog] has been found to have surface spotted degradation and discoloration.

Problems of Formulations of the Prior Art:-

Accordingly, it is understood from the foregoing that known formulations comprising alpha ketoanalogue of amino acids and other amino acids have been found to be unstable and inefficient for the treatment of chronic kidney failure particularly due to their decreased in-vitro dissolution and high degradation, more particularly under moisture and heat conditions, even more particularly during storage of the tablet form of the formulations comprising alpha ketoanalogue of amino acids.

Problem to be solved by the present invention:-

Therefore, the present invention aims to overcome above-described problems of the prior art by providing a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids which has been, surprisingly and unexpectedly, found to have increased in-vitro dissolution and controlled degradation, particularly under moisture and heat conditions, more particularly under moisture and heat conditions during storage of its tablet form, and hence, has been, surprisingly and unexpectedly, found to be protective against moisture and heat degradation, and having enhanced in-vitro dissolution meaning thereby expected to have enhanced in-vivo bioavailability of its active ingredients for efficient treatment of chronic kidney disease.

Objects of the Invention:-

The main object of the present invention is to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, and having increased in-vitro dissolution, and reduced and controlled degradation, particularly under moisture and heat conditions, more particularly under moisture and heat conditions during storage of its tablet dosages form.

The another main object of the present invention is to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, and having substantially increased in-vitro dissolution, and substantially reduced and controlled degradation, particularly under moisture and heat conditions, wherein its tablet dosages form is coated with moisture barrier coating composition.
The another object of the present invention is to provide a pharmaceutical
formulation comprising alpha ketoanalogue of amino acids and additional amino acids, and having possibility of enhanced *in-vivo* bioavailability of its active ingredients so as to achieve efficient treatment of chronic kidney disease.

This is also an object of the present invention to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, wherein formulation has reduced surface sensitivity.

This is also an object of the present invention to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, wherein formulation does not demonstrate surface spotted degradation and discoloration.

This is also an object of the present invention to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, wherein active ingredients of the formulation can get disintegrated from the formulation in minimum possible duration, preferably in less than 30 minutes.

This is also an object of the present invention to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, wherein active ingredients of the formulation can get disintegrated from the formulation in less than 30 minutes on contact with aqueous medium.

This is also an object of the present invention to provide a process for preparation of pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, wherein the formulation produced does not suffer from problems of sensitivity of alpha ketoanalogue of amino acids to heat and moisture.

The other objects and advantages of the present invention will become more apparent from the following description when read in conjunction with examples.

**Description and Preferred Embodiments of the Invention:**

With aim to provide a pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids, and having increased *in-vitro* dissolution, and reduced and controlled degradation, particularly under moisture and heat conditions, more particularly under moisture and heat conditions during storage of its tablet form, and having expected enhanced *in-vivo* bioavailability of its active ingredients so as to achieve efficient treatment of chronic kidney disease, the inventors have found that if croscarmellose sodium is used as disintegrant in combination with filler microcrystalline cellulose and binder polyvinyl pyrrolidone, the formulation comprising alpha ketoanalogue and additional amino acids, suprisingly and unexpectedly, demonstrates
increased *in-vitro* dissolution, and reduced arid controlled degradation under moisture and heat conditions even during its storage in the tablet dosage form.

The inventors have further found that if formulation comprising alpha ketoanologue of amino acids and additional amino acids, and croscarmellose sodium as disintegrant, microcrystalline cellulose as filler and polyvinyl pyrrolidone as binder is coated with moisture barrier coating composition comprising polyvinyl alcohol as film former, soya lecithin as plasticizer and solubilizing agent and xanthan gum as stabilizer and suspension agent, the *in-vitro* dissolution is, suprisingly and unexpectedly substantially increased, and degradation is, suprisingly and unexpectedly substantially reduced and controlled under the moisture and heat conditions even during its accelerated stability storage conditions as per ICH (International Conference on Harmonization) in the tablet dosage form.

Accordingly, the present invention relates to a pharmaceutical formulation having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions comprising alpha ketoanologue of amino acids and additional amino acids as active ingredients, and disintegrant, filler and binder, and coated with moisture barrier coating composition, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone, and moisture barrier coating composition comprises film former, plasticizer and solubilizing agent, and stabilizer and suspension agent, wherein film former is polyvinyl alcohol, plasticizer and solubilizing agent is soya lecithin, and stabilizer and suspension agent is xanthan gum.

Accordingly, in one embodiment, the present invention also relates to a pharmaceutical formulation having increased *in-vitro* dissolution, arid reduced and controlled degradation under moisture and heat conditions comprising alpha ketoanologue of amino acids and additional amino acids as active ingredients, and disintegrant, filler and binder, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone.

Therefore, the formulations of present invention have been found to have improved *in-vitro* dissolution, and reduced and controlled degradation of the drugs (the active ingredients) from the moisture and heat in the tested pack during their storage.

The inventors have also found that the formulations of present invention do not demonstrate surface spotted degradation and discoloration.

Further, with aim to overcome problems of sensitivity of alpha ketoanologue amino acids and additional amino acids to heat and moisture, the inventors have found that
if formulations of present invention are prepared by a dry granulation process while controlling the moisture absorption at every step of the process, it surprisingly and unexpectedly results in reduction of surface sensitivity of the tablet dosage form.

Accordingly, in another preferred embodiment, the present invention also relates to a process for preparation of pharmaceutical formulation for oral administration having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions comprising steps of mixing alpha ketoanalogue of amino acids and additional amino acids homogeneously with disintegrant, filler and binder followed by dry granulation of the resulted blend, and optionally step of lubrication followed by compressing the core to tablet form and then coating with moisture barrier coating composition comprising film former, plasticizer and solubilizing agent, and stabilizer and suspension agent, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone, and film former is polyvinyl alcohol, plasticizer and solubilizing agent is soya lecithin, and stabilizer and suspension agent is xanthan gum.

It may be noted that the formulations of present invention having substantially increased *in-vitro* dissolution, and substantially reduced and controlled degradation under the moisture and heat conditions even during its storage in the tablet dosage form are expected to have enhanced *in-vivo* bioavailability of its active ingredients.

Accordingly, in one of the embodiments of the present invention, the scope of the present invention also includes a pharmaceutical formulation having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions, and having enhanced *in-vivo* bioavailability of its active ingredients comprising alpha ketoanalogue of amino acids and additional amino acids as active ingredients, and croscarmellose sodium as disintegrant, microcrystalline cellulose as filler and polyvinyl pyrrolidone as binder.

In another preferred embodiment, the scope of the present invention also includes a pharmaceutical formulation having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions, and having enhanced *in-vivo* bioavailability of its active ingredients comprising alpha ketoanalogue of amino acids and additional amino acids as active ingredients, and croscarmellose sodium as disintegrant, microcrystalline cellulose as filler and polyvinyl pyrrolidone as binder, and coated with moisture barrier coating composition comprising polyvinyl alcohol as film former, soya
lecithin as plasticizer and solubilizing agent and xanthan gum as stabilizer and suspension agent.

In accordance with present invention, the alpha ketoanalogue of amino acids are selected from a group comprising calcium -3- methyl-2-oxo-valerate (alpha-ketoanalogue to isoleucine, calcium salt), calcium 4-methyl-2-oxo-valerate (alpha-ketoanalogue to leucine, calcium salt), calcium-2-oxo-3-phenylpropionate (alpha-ketoanalogue to phenylalanine, calcium salt), calcium -3- methyl-2-oxo butyrate (alpha-ketoanalogue to valine, calcium salt), and calcium-DL-2-hydroxy-4(methylthio)butyrate (alpha-hydroxyanalogue to methionine, calcium salt).

In accordance with present invention, the additional amino acids are selected from a group comprising L-lysine acetate, L-threonine, L-tryptophan, L-histidine, and L-tyrosine.

In accordance with one of the embodiments of the present invention, the filler is taken in an amount varying from about 12 to about 25% w/w of the uncoated formulation, preferably in an amount varying from about 15 to about 20% w/w of the uncoated formulation.

In accordance with one of the preferred embodiments of the present invention, it further comprises crospovidone as additional disintegrant.

In accordance with one of the embodiments of the present invention, the disintegrants comprising croscarmellose sodium and crospovidone are taken in an amount varying from about 4 to about 12% w/w of the uncoated formulation, preferably in an amount varying from about 8 to about 10% w/w of the uncoated formulation.

In accordance with one of the embodiments of the present invention, the croscarmellose sodium is employed as disintegrant in an amount varying from about 2 to about 6% w/w of the uncoated formulation, preferably in an amount varying from about 3 to about 5% w/w of the uncoated formulation.

In accordance with preferred embodiment of the present invention, the disintegrants croscarmellose sodium and crospovidone are taken in a weight ratio varying from about 1:1 to about 1:1.50, preferably about 1:1.25 in the uncoated formulation.

In accordance with one of the preferred embodiments of the present invention, the present formulation further comprises colloidal silicon dioxide as co-disintegrant and glidant.

In accordance with preferred embodiment of the present invention, the disintegrants, additional disintegrant and co-disintegrant being respectively croscarmellose
sodium, crospovidone and colloidal silicon dioxide are taken in an amount varying from about 8 to about 14% w/w of the uncoated formulation, preferably in an amount varying from about 9 to about 12% w/w of the uncoated formulation.

In accordance with one of the preferred embodiments of the present invention, the co-disintegrant colloidal silicon dioxide is taken in an amount varying from about 0.5 to about 2% w/w of the uncoated formulation, preferably in an amount varying from about 0.75 to about 1.2% w/w of the uncoated formulation.

In accordance with one of the preferred embodiments of the present invention, the binder is a combination of polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30, which are taken in an amount varying from about 5 to about 9% w/w of the uncoated formulation, preferably in an amount varying from about 6 to about 8% w/w of the uncoated formulation.

In accordance with one of the preferred embodiments of the present invention, the binder polyvinyl pyrrolidone K-90 is taken in higher amount than the binder polyvinyl pyrrolidone K-30. In accordance with one of the preferred embodiments of the present invention, the weight ratio of polyvinyl pyrrolidone K-90 to polyvinyl pyrrolidone K-30 preferably varies from about 1.10:1 to about 1.20:1. In accordance with more preferred embodiment of the present invention, the weight ratio of polyvinyl pyrrolidone K-90 to polyvinyl pyrrolidone K-30 is about 1.16:1.

In accordance with most preferred embodiment of the present invention, the core of the formulation tablet is produced by dry granulation, which is then given a coating of present formulation, which has been found to have capability of preventing the core from degradation on contact with environmental moisture and heat, and from degradation in the tested pack during storage. Accordingly, the presently provided formulation has been found to have capabilities to protect the drugs—the active ingredients from the moisture and heat, and to prevent the drugs—the active ingredients, from degradation during storage.

In accordance with most preferred embodiment of the present invention, the coating is mixture barrier coating composition comprising polyvinyl alcohol as film former, soya lecithin as plasticizer and solubilizing agent, and xanthan gum as stabilizer and suspension agent: In presently provided coated formulation, the soya lecithin has been found to act as plasticizer and solubilizing agent, and xanthan gum has been found to act as stabilizer and suspension agent.
The inventors have found that moisture barrier coating composition comprising polyvinyl alcohol, soya lecithin and xanthan gum enhances the release of drug from the core tablet.

In accordance with present invention, the coated formulation comprises about 5 to 7% of the moisture barrier coating composition.

In accordance with preferred embodiment of the present invention, moisture barrier coating composition comprises about 45.5% w/w of the polyvinyl alcohol as film former, about 2% w/w of the soya lecithin as plasticizer and solubilizing agent, about 0.5% w/w of the xanthan gum as stabilizer and suspension agent. The moisture barrier coating composition as used in present invention additionally comprises excipients selected from the group comprising talcum, titanium dioxide, quinoline yellow, opacifire and colouring agent amounting to about 52%.

With aim to overcome problems of surface discolouration, and degradation on exposure to heat and moisture, and poor dissolution of the formulation comprising alpha ketoanalogue of amino acid and amino acids, the inventors have found that coating compositions comprising hydroxy propyl methyl cellulose [HPMC], or hydroxyl ethyl cellulose [HEC], or sodium carboxy methyl cellulose [NaCMC], or ethyl cellulose [EC], or hydroxy propyl cellulose [HPC] as film former with other excipients including plasticizers, stabilizing agent and colourants did not show any reduction in surface discolouration and degradation on exposure to heat and moisture, and improvement in the in-vitro dissolution of the formulation.

The inventors made further efforts by initially seal coating the core tablet with instamoistshield of M/s. Ideal Cures which comprises polyvinyl alcohol in combination with polyethylene glycol as plasticizer followed by film coat with Opadry (Colorcon) containing hydroxypropylmethyl cellulose. The inventors found that even this coating composition did not provide sufficient and required moisture resistance and reduced discoloration.

After lot of research work, the inventors have found that it is only the coating composition comprising polyvinyl alcohol as film former in combination with soya lecithin as plasticizer and solubilizing agent and xanthan gum as stabilizer and suspension agent, which surprisingly and unexpectedly, results in a formulation having substantially increased in-vitro dissolution, and substantially reduced and controlled degradation under the moisture and heat conditions even during the accelerated stability storage conditions as per ICH (International Conference on Harmonization) in its tablet dosage form.
The inventors have found that only the combination of polyvinyl alcohol, soya lecithin and xanthan gum results in the best suitable moisture barrier composition, which provides an effective moisture barrier, reduces moisture entrapment on the surface at the same time allows the drugs to permeate through the coating more effectively.

In accordance with one of preferred embodiments of the present invention, the moisture barrier coating composition used in present invention is one available from Colorcon as Opadry AMB readymix.

In accordance with one of the preferred embodiments of the present invention, the core formulation further comprises lubricant. The lubricating agent is selected from the group comprising magnesium stearate and sodium starch fumarate, preferably the lubricating agent is magnesium stearate.

In accordance with one of the embodiments of the present invention, the filler may be selected from the group comprising microcrystalline cellulose, lactose, dicalcium phosphate, and mannitol. However, the inventors have found that when microcrystalline cellulose is used as filler in combination with other judiciously selected disintegrants and binders of present formulation, the present formulation demonstrates unexpectedly increased in-vitro dissolution, and unexpectedly reduced and controlled degradation.

In accordance with preferred embodiment of the present invention, microcrystalline cellulose is partially depolymerized cellulose, white, odorless and tasteless powder composed of porous particles having about 90% particles of particle size varying from about 70 to 130 micron, preferably of about 100 micron, and having moisture content less than 5%.

The inventors have found that it is the granular form of microcrystalline cellulose which is available as Avicel pH 102, surprisingly and unexpectedly, demonstrates better dissolution properties as compared to other grades of microcrystalline cellulose. Accordingly, the granular form of microcrystalline cellulose is most preferred in accordance with the present invention.

The inventors have also found that the judiciously selected above-described microcrystalline cellulose not only acts as filler, but also acts as a disintegrating agent due to its capillary action which gives bursting effect, thus lowering the disintegration time of the tablet on contact with aqueous medium. The presently selected microcrystalline cellulose has been found to have unique compressibility and carrying capacity which exhibits excellent properties as an excipient for the present formulation. It was found to compact well under minimum compression pressures and to have high binding capability.
and capable of resulting in extremely hard and stable core tablets, and yet disintegrate rapidly. The judiciously selected microcrystalline cellulose also demonstrates advantages of low friability, inherent lubricity, and highest diluting potential of all binders. Accordingly, it is believed, the judiciously selected microcrystalline cellulose acts as filler and as binder for the present formulation. The selected microcrystalline cellulose has been found to be highly absorptive due to its capillary action of its surface porosity, which indicates that it may also be acting as a carrier for liquids and yet retain free flowing and compression properties of the formulation.

In accordance with one of the preferred embodiments of the present invention, the disintegrant is combination of croscarmellose sodium and crospovidone. The inventors have found that when disintegrant crospovidone is combined with croscarmellose sodium as in present invention, the in-vitro dissolution of active ingredients is substantially increased.

In accordance with preferred embodiment of the present invention, the croscarmellose sodium having loss of moisture on drying \( \leq 10.0\% \) w/w, and pH of 5.0 to 7.0 in aqueous dispersion is preferred choice as it has been found effective in disintegrating large amounts of alpha ketoanalogure of amino acids and additional amino acids from the present tablet dosage form.

In accordance with preferred embodiment of the present invention, the crospovidone having loss of moisture on drying \( \leq 5.0\% \) w/w and pH of 5.0 to 8.0 in 1% w/v aqueous dispersion is preferred choice as it has been found effective in disintegrating large amounts of alpha ketoanalogure of aminp acids and additional amino acids from the present tablet dosage form.

In accordance with preferred embodiment of the present invention, the croscarmellose sodium is an internally crossed-linked sodium carboxymethyl cellulose, which due to its cross-linking has been found to have advantage of swelling in water, and as a result, the judiciously selected croscarmellose sodium has been found to provide superior drug disintegration and dissolution characteristics, and thereby improving the efficiency of the pharmaceutical formulation of present invention, which, accordingly, is expected to give better in-vivo bioavailability.

In accordance with one of the embodiments of the present invention, the binder may be selected from the group comprising carboxymethyl cellulose sodium, methyl cellulose, hydroxypropyl cellulose, and polyvinyl pyrrolidone. However, the inventors have found that when polyvinyl pyrrolidone is used as binder, particularly when
combination of polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30 is used as binder in combination with other judiciously selected filler and disintegrants of present formulation, the present formulation demonstrates unexpectedly increased in-vitro dissolution, and unexpectedly reduced and controlled degradation.

In accordance with preferred embodiment of the present invention, the combination of polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30 as binder has been found to enhance dissolution of the present formulation when used in combination with filler and disintegrant of the present invention.

In accordance with preferred embodiment of the present invention, the polyvinyl pyrrolidone-K-90 is one having molecular weight of about 100,000 and polyvinyl pyrrolidone-K-30 is one having molecular weight of about 50,000. The combination of these polyvinyl pyrrolidones has been found to act as binder and as well as dissolution enhancer in present formulation to enhance its in-vitro dissolution.

In accordance with present invention, the polyvinyl pyrrolidones are white hygroscopic powder with characteristic odor in contrast with most polymers which are readily soluble both in water as well as in a large number of organic solvents selected from the group comprising alcohol, amine, acid, amide, lactam. It has been found that due to increased solubility of polyvinyl pyrrolidones in aqueous medium, these serve as a dual phenomenon of binder as well as a dissolution enhancer in present formulation.

In accordance with one of the preferred embodiments of the present invention, the colloidal silicon dioxide used as co-disintegrant and glidant is the one having large specific surface area which has been found to have advantage of desirable flow characteristics which results in improvement of the flow properties of the present formulation thereby resulting in the formulation having advantage of absorbing moisture in the aqueous media to cause of swelling of the present formulation.

It may be noted that present pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids as active ingredients is non-ethanolic formulation.

The inventors have found that present formulation when comprising above-described ingredients in above-described ratio or relative amounts give more strength to the formulation in coated tablet dosage form resulting in the reduced friability.

It may be noted that above-described advantages of presently employed core excipients have been demonstrated by the inventors in the present formulation having presently provided coating of moisture barrier coating composition.
The inventors have observed that the present pharmaceutical formulation comprising alpha ketoanalogue of amino acids and additional amino acids as active ingredients, and croscarmellose sodium and crospovidone as disintegrants, microcrystalline cellulose as filler and polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30 as binders, and coated with moisture barrier coating composition comprising polyvinyl alcohol as film former, soya lecithin as plasticizer and solubilizing agent and xanthan gum as stabilizer and suspension agent, demonstrates substantially increased in-vitro dissolution, and is substantially stable under moisture and heat conditions during its storage in the tablet dosage form, and demonstrates disintegration of the formulation in minimum possible duration, preferably in less than 30 minutes, which, in view of its substantially increased in-vitro dissolution, and substantially reduced and controlled degradation are indicative of higher drug release accuracy in better consistency of dissolution and in desired drug quality.

Accordingly, it may also be noted that the formulations of present invention having substantially increased in-vitro dissolution, and substantially reduced and controlled degradation under the moisture and heat conditions even during its storage in the tablet dosage form are not only expected to have enhanced in-vivo bioavailability of its active ingredients, but are also expected to have improved patient compliance.

It may be noted that presently provided coated formulation may be used for treatment of chronic kidney disease, acute kidney disease, acute or chronic renal failure.

**Examples of the Invention:**

The present invention is now described with the help of following examples, which are not intended to limit scope of present invention, but have been incorporated for defining the best mode of performing the present invention.

For the experimental studies, five formulations were prepared in accordance with the present method of preparation as described hereinafore and having concentration of various ingredients as described and particularly provided in below Table - I, wherein Expt. 1, Expt. 2, Expt. 3, Expt. 4 are for comparative purpose and Expt. 5 is in accordance with most preferred embodiment of the present invention, which is included with the scope of present invention. The in-vitro dissolution, and degradation under the moisture and heat conditions were studied of all five formulations. It has been found that only the formulation of Expt. 5 demonstrated substantially increased in-vitro dissolution, and substantially reduced and controlled degradation under the moisture and heat conditions. The formulation of Expt. 5 was also subjected to stability storage test under accelerated
stability storage conditions as per ICH (International Conference on Harmonization) in its tablet dosage form and surprisingly and unexpectedly found to be substantially stable.

Table - 1

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Core Active Principles (API)</th>
<th>Composition of the tablet containing 630 mg dose of API (amount in mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Expt. 1</td>
</tr>
<tr>
<td>1.</td>
<td>Calcium -3- methyl-2-oxo-valerate</td>
<td>67</td>
</tr>
<tr>
<td>2.</td>
<td>Calcium 4 - Methyl-2-Oxovalerate</td>
<td>101</td>
</tr>
<tr>
<td>3.</td>
<td>Calcium - 2- oxo - 3-phenylpropionate</td>
<td>68</td>
</tr>
<tr>
<td>4.</td>
<td>Calcium -3- methyl-2- oxo butyrate</td>
<td>86</td>
</tr>
<tr>
<td>6.</td>
<td>L-Lysine Acetate</td>
<td>105</td>
</tr>
<tr>
<td>7.</td>
<td>L- Threonine</td>
<td>53</td>
</tr>
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<td>8.</td>
<td>L-Tryptophan</td>
<td>23</td>
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<tr>
<td>9.</td>
<td>L-Histidine</td>
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<tr>
<td>10.</td>
<td>L- Tyrosine</td>
<td>30</td>
</tr>
</tbody>
</table>

Core Excipients

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Core Excipients</th>
<th>Amount in mg</th>
</tr>
</thead>
<tbody>
<tr>
<td>11.</td>
<td>Microcrystalline Cellulose</td>
<td>140.00 80.00 125.00 180.00 180.00</td>
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<tr>
<td>12.</td>
<td>Crospovidone</td>
<td>25.00 30.00 40.00 50.00 50.00</td>
</tr>
<tr>
<td>13.</td>
<td>Sodium starch glycollate</td>
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<td>Polyvinyl Pyrrolidone -30</td>
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<td>Colloidal silicon dioxide</td>
<td>12.00 12.00 8.00 11.00 11.00</td>
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<td>Talc</td>
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<td>Magnesium Stearate</td>
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<td>Powder Cellulose</td>
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<td>-- -- -- 35.00 35.00</td>
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<tr>
<td>21.</td>
<td>Croscarmellose Sodium</td>
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Coating Material

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<th>Amount in mg</th>
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<td>22.</td>
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<td>27 27 27 27 --</td>
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<tr>
<td>23.</td>
<td>Opadry (Film coating)</td>
<td>27 27 27 18 --</td>
</tr>
<tr>
<td>24.</td>
<td>Opadry aqueous moisture barrier</td>
<td>-- -- -- -- 65</td>
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The inventors confirm that it is only the formulation of Expt 5, which demonstrated substantially increased in-vitro dissolution, and substantially reduced and controlled degradation under the moisture and heat conditions, and even surface spotted degradation and discoloration was not observed in coated tablets formed from this formulation confirming their reduced surface sensitivity, and hence, it can be concluded that coated tablets formed from formulation of Expt. 5 overcome problems of sensitivity of alpha ketoanalogue of amino acids and additional amino acids to heat and moisture even during storage, and demonstrate substantially increased in-vitro dissolution.

The inventors also confirm that during the in-vitro studies the active ingredients of the formulation of Expt. 5 get disintegrated from the formulation in less than 30 minutes on contact with aqueous medium.

The inventors also confirm that coated tablets coated with Instamoistshield or with Opadry film coating did not demonstrate increased in-vitro dissolution, and controlled degradation under the moisture and heat conditions.

The in-vitro dissolution (%) results of formulation of Expt. 5 were compared with known formulation available under the name Ketosteril using the dissolution apparatus USP, dissolution media purified water at RPM 75 for 45 min at 25°C under 60% relative humidity [RH] and at 40°C under 75% RH, and the data is presented in Table - II, which confirms that the in-vitro dissolution of formulation of Expt. 5 is far superior than the formulation available under the name Ketosteril.
<table>
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<th>at 40°C under 75% RH</th>
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<td>Formulation of Expt. 5</td>
<td>Ketosteril</td>
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<td>Ca-DL-2Hydroxy 4 methyl thiobutyrate (Methionine)</td>
<td>99</td>
<td>32</td>
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<tr>
<td>Ca-3 Methyl-2-oxo butyrate (Valine)</td>
<td>98</td>
<td>32</td>
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<td>Ca-4 Methyl-2-oxo-valerate (Leucine)</td>
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<td>30</td>
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<td>Ca-3 Methyl-2-oxo-valerate (Isoleucine)</td>
<td>98</td>
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<td>Ca-2-oxo-3 phenylpropionate (Phenylalanine)</td>
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<td>L-Tryptophan</td>
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<td>L-Threonine</td>
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<td>L-Histidine</td>
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<td>23</td>
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<tr>
<td>L-Lysine Acetate</td>
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<td>20</td>
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Claims

1. A pharmaceutical formulation having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions comprising alpha ketoanalogue of amino acids and additional amino acids as active ingredients, and disintegrant, filler and binder, and coated with moisture barrier coating composition, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone, and moisture barrier coating composition comprises film former, plasticizer and solubilizing agent, and stabilizer and suspension agent, wherein film former is polyvinyl alcohol, plasticizer and solubilizing agent is soya lecithin, and stabilizer and suspension agent is xanthan gum.

2. A pharmaceutical formulation as claimed in claim 1, wherein said alpha ketoanalogue of amino acids are selected from a group comprising calcium -3-methyl-2-oxo-valerate (alpha-ketoanalogue to isoleucine, calcium salt), calcium 4-methylJ-2-oxo-valerate (alpha-ketoanalogue to leucine, calcium salt), calcium-2-oxo-3-phenylpropionate (alpha-ketoanalogue to phenylalanine, calcium salt), calcium -3- methyl-2-oxo butyrate (alpha-ketoanalogue to valine, calcium salt), and calcium-DL-2-hydroxy-4(methylthio)butyrate (alpha-hydroxyanalogue to methionine, calcium salt).

3. A pharmaceutical formulation as claimed in any one of claims 1 to 2, wherein said additional amino acids are selected from a group comprising L-lysine acetate, L-threonine, L-tryptophan, L-histidine, and L-tyrosine.

4. A pharmaceutical formulation as claimed in any one of claims 1 to 3, wherein said filler is taken in an amount varying from about 12 to about 25% w/w of the uncoated formulation, preferably in an amount varying from about 15 to about 20% w/w of the uncoated formulation.

5. A pharmaceutical formulation as claimed in any one of claims 1 to 4, wherein said formulation further comprises additional disintegrant, which is crospovidone.

6. A pharmaceutical formulation as claimed in any one of claim 5, wherein said disintegrants comprising croscarmellose sodium and crospovidone are taken in an amount varying from about 4 to about 12% w/w of the uncoated formulation, preferably in an amount varying from about 8 to about 10% w/w of the uncoated formulation.
7. A pharmaceutical formulation as claimed in any one of claims 1 to 6, wherein said croscarmellose sodium and crospovidone are taken in a weight ratio varying from about 1:1 to about 1:1.50, preferably about 1:1.25 in the uncoated formulation.

8. A pharmaceutical formulation as claimed in any one of claims 1 to 7, wherein said croscarmellose sodium is taken in an amount varying from about 2 to about 6% w/w of the uncoated formulation, preferably in an amount varying from about 3 to about 5% w/w of the uncoated formulation.

9. A pharmaceutical formulation as claimed in any one of claims 1 to 8, wherein said formulation further comprises co-disintegrant and glidant which is colloidal silicon dioxide.

10. A pharmaceutical formulation as claimed in claim 9, wherein said disintegrants, additional disintegrant and co-disintegrant are taken in an amount varying from about 8 to about 14% w/w of the uncoated formulation, preferably in an amount varying from about 9 to about 12% w/w of the uncoated formulation.

11. A pharmaceutical formulation as claimed in either of claim 9 or 10, wherein said co-disintegrant is taken in an amount varying from about 0.5 to about 2% w/w of the uncoated formulation, preferably in an amount varying from about 0.75 to about 1.2% w/w of the uncoated formulation.

12. A pharmaceutical formulation as claimed in any one of claims 1 to 11, wherein said binder is a combination of polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30.

13. A pharmaceutical formulation as claimed in claim 12, wherein said polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30 are taken in an amount varying from about 5 to about 9% w/w of the uncoated formulation, preferably in an amount varying from about 6 to about 8% w/w of the uncoated formulation.

14. A pharmaceutical formulation as claimed in any one of claims 12-13, wherein said polyvinyl pyrrolidone K-90 is taken in higher amount than polyvinyl pyrrolidone K-30.

15. A pharmaceutical formulation as claimed in claim 14, wherein weight ratio of polyvinyl pyrrolidone K-90 to polyvinyl pyrrolidone K-30 varies from about 1.10:1 to about 1.20:1, preferably it is about 1.16:1.

16. A pharmaceutical formulation as claimed in any one of claims 1-15, wherein said formulation further comprises lubricant.
17. A pharmaceutical formulation as claimed in claim 16, wherein said lubricating agent is magnesium stearate.

18. A pharmaceutical formulation as claimed in any one of claims 1-17, wherein said microcrystalline cellulose is partially depolymerized cellulose composed of porous particles having about 90% particles of particle size varying from about 70 to 130 micron, preferably of about 100 micron, and having moisture content less than 5%.

19. A pharmaceutical formulation as claimed in any one of claims 1-18, wherein said macrocrystalline cellulose is granular form of microcrystalline cellulose.

20. A pharmaceutical formulation as claimed in any one of claims 1-19, wherein said croscarmellose sodium has loss of moisture on drying $\leq 10.0\%$ w/w, and pH of 5.0 to 7.0 in aqueous dispersion.

21. A pharmaceutical formulation as claimed in any one of claims 1-20, wherein said crospovidone has loss of moisture on drying $\leq 5.0\%$ w/w and pH of 5.0 to 8.0 in 1% w/v aqueous dispersion.

22. A pharmaceutical formulation as claimed in any one of claims 1-21, wherein said croscarmellose sodium is internally crossed-linked sodium carboxymethyl cellulose.

23. A pharmaceutical formulation as claimed in any one of claims 1-22, wherein said polyvinyl pyrrolidone-K-90 has molecular weight of about 10,00,000 and polyvinyl pyrrolidone-K-30 has molecular weight of about 50,000.

24. A pharmaceutical formulation as claimed in any one of claims 1-23, wherein said formulation comprises alpha ketoanalogue of amino acids and additional amino acids as active ingredients, and disintegrant is combination of croscarmellose sodium and crospovidone, filler is microcrystalline cellulose, and binder is combination of polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30, and is coated with moisture barrier coating composition comprising film former, plasticizer and solubilizing agent, and stabilizer and suspension agent, wherein film former is polyvinyl alcohol, plasticizer and solubilizing agent is soya lecithin, and stabilizer and suspension agent is xanthan gum.

25. A pharmaceutical formulation as claimed in any one of claims 1-24, wherein coated formulation comprises about 5 to 7% of the moisture barrier coating composition.

26. A pharmaceutical formulation as claimed in any one of claims 1-25, wherein said coating composition comprises about 45.5% w/w of the polyvinyl alcohol as film.
former, about 2% w/w of the soya lecithin as plasticizer and solubilizing agent, about 0.5% w/w of the xanthan gum as stabilizer and suspension agent.

27. A pharmaceutical formulation as claimed in any one of claims 1-26, wherein said coating composition additionally comprises excipients selected from the group comprising talcum, titanium dioxide, quinoline yellow, opacifire and colouring agent amounting to about 52%.

28. A pharmaceutical formulation having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions comprising alpha ketoanalgue of amino acids and additional amino acids as active ingredients, and disintegrant, filler and binder, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone.

29. A pharmaceutical formulation as claimed in claim 28, wherein said disintegrant additionally comprises crospovidone, and said binder is combination of polyvinyl pyrrolidone K-90 and polyvinyl pyrrolidone K-30.

30. A pharmaceutical formulation as claimed in any one of claims 1-29, wherein said formulation is non-ethanolic formulation.

31. A process for preparation of pharmaceutical formulation of any one of claims 1-27, 30 for oral administration having increased *in-vitro* dissolution, and reduced and controlled degradation under moisture and heat conditions comprising steps of mixing alpha ketoanalgue of amino acids and additional amino acids homogeneously with disintegrant, filler and binder followed by dry granulation of the resulted blend, and optionally step of lubrication followed by compressing the core to tablet form and then coating the resulted tablet form with moisture barrier coating composition, wherein disintegrant is croscarmellose sodium, filler is microcrystalline cellulose and binder is polyvinyl pyrrolidone, and moisture barrier coating composition comprises polyvinyl alcohol as film former, soya lecithin as plasticizer and solubilizing agent and xanthan gum as stabilizer and suspension agent.

32. A pharmaceutical formulation substantially as herein described with reference to foregoing examples.

33. A process for preparation of pharmaceutical formulation substantially as herein described with reference to foregoing examples.
**INTERNATIONAL SEARCH REPORT**

A. CLASSIFICATION OF SUBJECT MATTER

INV. A61K9/28 A61K31/00

ADD.

According to International Patent Classification (IPC) into both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

EPO-Internal, CHEM ABS Data, EMBASE, WPI Data, BIOSIS, FSTA

C. DOCUMENTS CONSIDERED TO BE RELEVANT

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Further documents are listed in the continuation of Box C. See patent family annex.

Date of the actual completion of the international search

16 April 1 2012

Date of mailing of the international search report

25/04/2012

Name and mailing address of the ISA

European Patent Office, P.B. 5818 Patentlaan 2
NL - 2280 HV Rijswijk
Tel. (+31-70) 340-2040, Fax: (+31-70) 340-3016

Authorized officer

Schul e, Stefani e

Form PCT/ISA/210 (second sheet) (April 2005)
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