CALCIUM PIDOLATE

To a considerable extent, calcium is incorporated in the skeleton. In metabolic respects, calcium is also involved in the body's most vital functions. It thus plays a key role in growth disorders: decalcification, delayed weight and height gain, rickets, loss of appetite, etc., and is at the centre of the physiological progression of bony tissue towards osteoporosis.

Finally, calcium supplementation is indicated in all fracture consolidation disorders: delayed consolidation, malunion, pseudoarthrosis, etc. But calcium is also the key to metabolic exchanges in cells and nerves, providing essential support in maintaining a nutritional balance. Calcium has been used as a treatment for many years, but has not always proved effective. After all, there's no point in absorbing large quantities of a calcium mineral that is eliminated straight away.

*Calcium Pidolate is the only drug in which the calcium is transported by a protein substrate that is directly assimilable and specific to the body’s bony matrix.*

Thanks to this original property, Calcium Pidolate can help the body's own calcium in performing its two primary roles:

- in any adverse changes in bony tissue during its growth, during bone ageing or traumatic bone lesions, *Calcium Pidolate can provide both a protein framework and a mineral source,*
- as regards various metabolic functions of calcium, recent studies employing labelled Calcium Pidolate have demonstrated that the substance is perfectly absorbed and distributed throughout various tissues and that it *stimulates numerous hormonal and metabolic activities.*

ROLE OF CALCIUM IN THE BODY

BONE CALCIUM

The skeleton, with its 208 bones, is the most important component of the locomotor apparatus. Moreover, bones protect the brain, the spinal cord and the sensory organs. A long bone comprises two parts: the slender, central tubular section (the shaft) simply consists of a thick layer of very compact, hard bone known as cortical bone. At the two ends of the bone (or epiphyses) the bony substance is spongy, trabecular and less resistant.

Bone structure

Bone is formed from a framework, or protein matrix of collagen tissue. Collagen, in turn, is formed from fibroblasts, which reach maturity when an amino acid, proline, appears in their midst. Collagen itself appears when the proline is transformed into hydroxyproline. These amino acids receive *calcium and phosphorus,* substances that bind to the acids and give bone its solidity and rigidity. This mineral mix contains about twice as much calcium as phosphorus.

![normal bone structure](image)
Bone remodeling

Bone is always in a state of constant change. Certain cells known as osteoclasts hollow out resorption cavities in the bone tissue, eliminating the protein matrix and its mineral content. Other cells known as osteoblasts gradually fill in these resorption cavities and use proline to construct the protein framework. Calcium and phosphate salts then arrive and bind themselves to this framework. These changes play a metabolic role and, particularly during growth, perform morphological and structural functions. They are regulated by various hormones. Parathyroid hormone, or parathormone (PTH), promotes bone resorption but also increases the intestinal absorption of calcium in the presence of vitamin D. Thyrocalcitonin (TCT), from the thyroid, inhibits bone resorption but slightly increases the intestinal absorption and urinary elimination of calcium. Growth remains under the control of growth hormone (GH).

THE ORIGINAL PROPERTIES OF CALCIUM PIDOLATE

![Chemical structures]

In Calcium Pidolate the calcium ion is bound to two molecules of pyrrolidone carboxylic acid that act as protein supports. None of the calcium salts currently employed therapeutically (chloride, carbonate, lactate, gluconate, glucoheptonate, gluconolactate, gluconagalactogluconate, lactobionate, etc.) contain a protein support. Consequently:

**Calcium Pidolate is the only drug in which the calcium possesses a protein support.**

Proteins are the most important constituents of tissue cells in the body and are involved in the most vital functions. Additionally, a drug that possesses protein constituents can be guaranteed to benefit from good gastrointestinal absorption, easy circulation in the blood and excellent tissue binding. In the case of Calcium Pidolate the calcium is much more readily liberated and ionized than in any other non-protein calcium salt.

Finally, we know that bone forms a framework or matrix. This framework is protein in nature. The collagen tissue of which it is composed is formed from fibroblasts. These reach maturity when proline appears in their midst. And collagen itself appears when the proline is transformed into hydroxyproline. These are the amino acids that receive calcium and phosphorus.

One simply has to look at the close chemical relationship between pyrrolidone carboxylic acid (PCA), proline and hydroxyproline to realize that:

**Calcium Pidolate is the only drug whose protein support is specific to the body’s bony framework.**
CALCIUM DEFICIENCIES GROWTH DISORDERS

Growth disorders and decalcification are just as common in western countries, as a result of a highly processed diet, inadequate exposure to the sun or insufficient breast-feeding, as in countries whose economies are still developing and whose populations may suffer from an inadequate or unbalanced diet.

The population migrations that are such a common feature of the modern world, with the sudden consequent changes in diet, are also relevant, as is the practice of bottle-feeding. Nevertheless, the foetal skeleton develops, particularly during the last two months of pregnancy, at the expense of the maternal stock of calcium and phosphate. This explains why women with multiple pregnancies are more likely to suffer from osteoporosis at an earlier age.

Decalcification

A decrease in calcium levels in the body can result in shortening of the bones and reduction in tooth height, bony fragility and multiple caries. This shortage of calcium may involve a reduction in blood calcium levels or hypocalcaemia. This condition is often accompanied by nervous disorders: sensory impairment, forced extension of the fingers in “obstetrician’s hand”, and possibly even tetanic crises.

This delayed growth is often associated with educational retardation in children or prepubertal adolescents if the dietary and hygienic conditions are less than ideal at an age when growth needs are pressing. This malnutrition and associated adjustment difficulties are also currently observed in many adolescents as a result of the veritable “dietary anarchy” that seems so fashionable these days.

Even certain overworked adults who are unable to devote sufficient time to their diet, or young, highly-strung women who follow excessively restrictive diets can find themselves exposed to calcium deficiency and hypocalcaemia, with consequent tetanic crises. While these are not genuinely serious in themselves, they are always disturbing for the victims and those around them.

CALCIUM PIDOLATE AND GROWTH DISORDERS

121 cases involving growth disorders have been described in five clinical studies: 1, 16, 33, 34, 38. These conditions affected all ages, from infants to older children, and involved the following:

- rickets 59 cases
- - anorexia, thinness, retarded weight and height gain, behavioural or sleeping disorders 41 cases
- premature infants 14 cases
- protein depletion 3 cases
- retarded dentition 3 cases
- tetany 1 case

Some of these children were treated with a syrup containing the combination of Calcium Pidolate (2.5 g/100 ml) and lysine pidolate (5 g/100 ml). We know the part that protein depletion plays in various growth retardation conditions in children and it seemed desirable to combine the specific calcium protein therapy of Calcium Pidolate with significant supplementation of one of the eight essential amino acids.
Overall results

Rickets

Rickets results from a deficiency of vitamin D, a poor diet or a lack of sunshine. It is often observed before the age of two years, and occasionally in later life. A pubertal form also exists. The bones in the skull remain soft, resulting in delayed closure of the fontanelle. Swelling of the costal cartilage (rachitic rosary) and joints with, subsequently, curvature of long bones are observed.

Significant supplies of vitamin D combined with regular calcium therapy are indicated. If hypocalcaemia is present, or if the child suffers from tetanic crises, the calcium therapy will even need to precede the vitamin treatment.

Preventive treatment is always the wiser option. Growth will need to be monitored every three months, almost up until puberty.

Chronic malnutrition

Chronic malnutrition can occur at any age and in all societies, and can lead to thinness, growth retardation, hypotonicity, hypoproteinaemia, calcium deficiency, anaemia, hypovitaminosis and mucosal atrophy. It can even be accompanied by nutritional oedema (e.g. due to protein undernutrition), diarrhoea and dehydration.

Frequently the absence of milk and dairy products (cheese, etc.) in the diet results in both calcium and protein deficiency.

CALCIUM PIDOLATE AND OSTEOPOROSIS

Bone is in a state of constant change. Osteoporosis can be defined as the predominance of resorption over formation with a consequent overall loss in bone mass. The salient feature of osteoporosis, a disease of "porous bones", is a general deficiency of bony tissue, both as regards the protein framework and calcium content.
The risk of fracture

It is the spongy, trabecular bone that is most likely to give way, whether in the limbs (particularly the neck of femur) or the vertebral bodies.

The anterior section of the vertebral body possesses an area of least resistance at which point the vertebra collapses and assumes the shape of a corner or "wedged vertebra". The upper extremity of the femur is a cantilevered structure that supports the weight of the trunk, the head and upper limbs. Despite the bone span arrangement, a "fractured neck of femur" can occur at various sites, highlighted above in bold lines.

Determination of the fracture threshold

Nowadays, bone mass can be measured precisely by photon densitometry. The bones of the forearm are placed in front of a photon source. A scintillation counter then measures the bone mineral content and the width of the bone. The mineral index is an expression of the relationship between these two parameters and reflects the density of the bony tissue. A fracture risk threshold has now been defined on the basis of numerous measurements.

Fracture risk ≤ a mineral index of 0.50

CALCIUM PIDOLATE AND OSTEOPOROSIS

The treatment of various forms of osteoporosis with Calcium Pidolate has been reported in five clinical studies: 9, 10, 20, 29, 38. The cases of osteoporosis were subdivided into:

- post-menopausal osteoporosis 25 cases
- osteoporosis after immobilization 15 cases
- common osteoporosis 14 cases

and:

- iatrogenic osteoporosis, including:
  - post-corticosteroid therapy and bismuth-induced osteoporosis 6 cases
  - osteoporosis + arthropathies, including
    - association with osteoarthritis, rheumatoid arthritis and ankylosing spondylitis 4 cases
  - osteoporosis + osteopathies, including:
    - association with Paget's disease, osteomalacia, spasmodic back pain, juvenile epiphysitis (Scheuermann disease), bone metastases and vertebral lysis 12 cases
In most of the above cases Calcium Pidolate was used curatively. It is self-evident that, in such chronic conditions, treatment should persist for a long time if the aim is to maintain any improvement. Moreover, certain patients with an increased fracture risk (multipara after the menopause for example) can benefit from regular preventive treatment.

**Overall results**

![Pie chart showing distribution: 74% Excellent and good, 20% Average, 6% No improvement]

**OSTEOPOROSIS**

- **Osteoporosis according to age. The mineral content decreases with age**
  - Slowly and regularly in men, but only from the age of 65 to 70 years;
  - Rapidly in women, from the menopause, i.e. 15 to 25 years earlier.
  - Age-related bone loss has been estimated at 0.3/0.7 % of bone mass per year, increasing to 1 to 3 % per year in osteoporosis.

- **Osteoporosis according to sex**

  The bone mineral content in women is always lower than that in men, whether compact bone (shaft) or spongy bone in the limbs is involved. The disappearance of hormonal activity around the age of 50 means that more than a quarter of women are at risk of suffering osteoporosis in the decade following the menopause: this is known as post-menopausal osteoporosis. The hormones most frequently implicated are the oestrogens which, while the ovaries are secreting, curb bone resorption. This explains why hormone replacement therapy is commonly prescribed, in association with calcium treatment, when menstruation ceases. Moreover, each pregnancy depletes a woman's bone stock. Successive pregnancies accelerate this trend and a multipara can reach the fracture threshold ten years earlier than a nullipara. It is therefore essential to monitor the mother's skeleton after each pregnancy.
CALCIUM PIDOLATE AND OSTEOPOROSIS

Experimental anti-osteoporotic activity

Professor F. BERTE (15) induced osteoporosis in rats by placing an elastic ligature around the femoro-tibial joint. A control group was left untreated. One group was treated with calcium gluconolactobionate and another with Calcium Pidolate, orally, at an equimolar dose of 135 mg/kg Ca++ per day.

Osteoporotic index

The group treated with Calcium Pidolate showed a much lower osteoporotic index than the group treated with calcium gluconolactobionate and the untreated control group.

FRACTURE CONSOLIDATION

Conditions for good consolidation

A fracture should be reduced as accurately as possible, either manually or surgically by internal fixation. Considerable progress has been made in the development of corresponding implants: pins, nails, plates, screws, etc.. used both provisionally and permanently. The reduced fracture should be fixed as effectively as possible, and in most cases this is achieved with a plaster cast.

Consolidation periods

The average consolidation periods in adults are shown in the following table. The age of the patient should also be taken into account: fractures consolidate faster in children than in adults and faster in adults than in elderly patients. Fractures of the lower limb take longer to consolidate in obese patients compared to thin patients.

<table>
<thead>
<tr>
<th>Bone</th>
<th>Area</th>
<th>Consolidation period</th>
</tr>
</thead>
<tbody>
<tr>
<td>Humerus: shaft:</td>
<td>1 to 3 months</td>
<td></td>
</tr>
<tr>
<td></td>
<td>extremities:</td>
<td>2 to 3 months</td>
</tr>
<tr>
<td>Radius: lower extremity:</td>
<td>1 to 1½ months</td>
<td></td>
</tr>
<tr>
<td>2 bones in the forearm:</td>
<td>3 to 4 months</td>
<td></td>
</tr>
<tr>
<td>Femur: neck or shaft:</td>
<td>3 to 5 months</td>
<td></td>
</tr>
<tr>
<td>Tibia:</td>
<td>3 months</td>
<td></td>
</tr>
<tr>
<td>2 bones in the leg:</td>
<td>3 to 4 months</td>
<td></td>
</tr>
<tr>
<td>2 malleoli in the ankle:</td>
<td>2 to 3 months</td>
<td></td>
</tr>
</tbody>
</table>

CALCIUM PIDOLATE AND FRACTURES
The prevention or treatment of delayed consolidation of single and multiple fractures has been investigated in six clinical trials: 6, 10, 23, 29, 36, 37. The following were investigated in particular:

- **closed single fractures** 46 cases
  including fractures of the neck of femur
- **closed double fractures** 20 cases
  (fractures of the two bones in the leg treated by intramedullary nailing)
- **multiple fractures, in most cases** 20 cases
  in patients with severe polytrauma
- **open fractures** 12 cases

The following were also studied:

- **pseudoarthrosis** 25 cases
- **decalcifying algodystrophy** 10 cases

The rate of consolidation was the specific subject of study in a comparison of two series of twenty patients each, one series forming the control group and the other treated with Calcium Pidolate. While callus thickness was "almost doubled", the diagram on page 11 makes it clear that the time to onset of callus formation was not doubled but rather shortened (a doubling of this time would, in fact, be a disadvantage of any treatment).

This study demonstrated (see below: Weight bearing of leg fractures) that, from the second month, the time to onset of callus formation was shortened and callus thickness was almost doubled in the treated group compared to the control group.

### Overall results for fractures

![Pie chart showing distribution of outcomes for fractures: 14.5% Excellent and good, 24.5% Average, 60% No improvement.]
DELAYED CONSOLIDATION

The delay can be expressed in clinical terms:

- by pain on mobilization,
- by pain on commencement of weight-bearing,
- by abnormal mobility during examination,
- by signs of inflammation: hot, swollen, inflamed skin around the fracture site;

and in radiological terms:

- by a clearly discernible gap between the two fragments,
- by calculus that is not very thick or dense,
- by a prolongation of the time to onset of callus formation,
- by abnormally decalcified bone extremities.

Malunion

Malunion is when a consolidated fracture or a fracture undergoing calcification is poorly positioned, with overlapping or rotation of the fragments, etc. Surgical correction is generally required.

Pseudoarthrosis

In this condition the bony callus has not formed. The fragments remain mobile and a "pseudo-joint" is formed at the fracture site. This results from failure of the protein framework of callus to retain its calcium content. Mobility of the fragments persists and, in the long term, this often becomes painless. The x-ray image is identical to that of delayed consolidation. Furthermore, the bone ends are considerably widened, resembling "elephant feet".

CALCIUM PIDOLATE AND FRACTURES

Weight-bearing of leg fractures

F. VIVES et al. (37) investigated two comparable groups of 20 fractures each. In all cases these were closed fractures of the two leg bones (in the mid-third) treated by intramedullary nailing. One group received 0.405 g/day of Calcium Pidolate while the other, control, group received no calcium therapy. These results were assessed according to two criteria:

- thickness of callus
- time to onset of callus formation

These very clear differences allowed earlier weight-bearing
METABOLIC CALCIUM

Calcium is the most abundant mineral in the body. Not only as a result of its predominance in the skeleton, but because it is also present throughout the body, which can be divided into two worlds: an intracellular world, which involves all the activities and reactions that occur in each cell of our tissues and an extracellular world made up of blood, lymph and interstitial fluid that bathes the cells. The cellular membrane - a genuine iron curtain - separates these two worlds.

In the past the cellular membrane used to be thought of merely as a kind of film. We now know, however, that the smallest single-celled creature is surrounded by an impressive barrier consisting of four to six layers of different molecules, all possessing considerable biological activity.

Calcium is present in the membrane, the intracellular compartment and the extracellular compartment.

And it is calcium that controls the permeability of the membrane, i.e. all exchanges between the two major sectors of the body.

Calcium, the key to exchanges

Greatly oversimplifying, it can be stated that no activity begins in a cell before a certain amount of potassium has flowed out of the cell and has been replaced by a quantity of sodium flowing in from the extracellular compartment; and that this activity will only cease when the same exchange in the opposite direction has taken place. Calcium is the substance that opens or closes the transmembranous channel gates (in the true meaning of the term).

CALCIUM PIDOLATE AND METABOLISM

Humoral recalcification

J.M. GAZAVE (13) induced severe, fatal hypocalcaemia with a s.c. injection of 1.5 g/kg of sodium EDTA (ethylene diamine tetraacetate), a pointer to blood calcium levels. He studied three groups of twenty rats: a control group, a group treated with calcium glutamate and a group treated with Calcium Pidolate with i.p. injections of an equimolar dose of 45 mg/kg Ca++. The following percentages were measured:

Grafik einfügen
Survival percentages for the various treatments
Recalcification of an experimental matrix

L. ROBERT et al. (32) (Connective Tissue Laboratory - Henri Mondor University Hospital - Créteil - Paris) studied bone matrices in rabbits and subsequently prepared an experimental model of a matrix that could be calcified by a homogenate of collagen fibres and used this matrix to incubate five different calcium salts at an equimolar concentration of 3.75 mm Ca++. The titrations were performed using a scintillation counter (number of counts per minute = cpm). The following substances were investigated: calcium acetate, calcium lactate, calcium gluconate, calcium chloride and Calcium Pidolate.

Radioactivity of incubation supernatants in relation to the time measured for fibres of insoluble collagen. The more the calcium salt persists in the supernatant, the less it binds to the matrix. Accordingly, the most effective salts were Calcium Pidolate and calcium lactate.

METABOLIC CALCIUM

Calcium and the control of cellular activity

Because of its central effect on the membrane, calcium modulates the activity of every cell. Each time a cell performs its function, its membrane is the site of changes in polarity corresponding to transmembranous exchanges of sodium and potassium. These changes in polarity are expressed in numerous ways:

- the conduction of neural impulses along the nerves
- the secretion of chemical mediators: at the nerve endings,
- the effect of the neural impulses is prolonged by the effect of numerous "neurological hormones", e.g. adrenaline, acetylcholine, histamine, serotonin, dopamine. These are also known as neurotransmitters.

All of these secretions depend upon calcium:

- the activity at the neuromuscular junction,
- the muscular contraction of the voluntary or involuntary muscles: heart, blood vessels, bronchi, intestine, etc.
Calcium and nutritional balance

Calcium also intervenes in numerous bodily functions:

- **the body's defences**: without calcium the white blood cells are unable to intercept antigens, aggressors against the individual.
- **nutritional processing**: all the substances that are constantly being manufactured or broken down in the body do so under the influence of enzymes associated with mineral salts, primarily with calcium.
- **clotting**: calcium intervenes on at least 6 different levels in the series of chain reactions resulting in the formation of a clot, the body's principal defence against bleeding.
- **vitamins**: vitamins A, B, C, D and K can only carry out their functions in the presence of calcium. Vitamin D, the "anti-rickets" vitamin, is known to be important in fixing the calcium content.

### CALCIUM PIDOLATE AND METABOLISM

**Secretion of hormones involved in calcification**

P. FRANCHIMONT et al. (11) (Radioimmunology Laboratory, Institute of Medicine, Liège - Belgium) conducted a double-blind study with 10 healthy volunteers to investigate the effects of an inorganic salt, calcium chloride, and Calcium Pidolate on the hormones that promote, or reduce, calcification.

<table>
<thead>
<tr>
<th>Parameters investigated</th>
<th>Significance of changes with Calcium chloride</th>
<th>Calcium Pidolate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Increase in the level of GH</td>
<td>not significant</td>
<td>p&lt;0.01</td>
</tr>
<tr>
<td>Increase in the level of calcitonin</td>
<td>not significant</td>
<td>p&lt;0.01</td>
</tr>
<tr>
<td>Decrease in the level of parathormone</td>
<td>not significant</td>
<td>p&lt;0.05</td>
</tr>
</tbody>
</table>

Only Calcium Pidolate produced a statistically significant effect on the three main calcification hormones: it increased GH and calcitonin levels and decreased levels of parathormone, the substance that induces bone resorption.

**Effects on calcaemia, calciuria and hydroxyprolinuria**

Professor P. BENOIT, Associate Professor F.X. MICHELET et al. (4) (Institute of Stomatology - Bordeaux - France) conducted a comparative study that followed up 2 equal groups of 25 patients for three weeks after maxillofacial surgery:

- traditional oral calcium therapy of 2.5 g elemental calcium per day in one group
- oral Calcium Pidolate at the dosage of 0.405 g elemental calcium per day in the other group (i.e. corresponding to a dose of Ca++ six times smaller).
The following were measured in relation to the mean value before treatment:

- calcaemia

<table>
<thead>
<tr>
<th></th>
<th>Calcium Pidolate 0.405 g/d</th>
<th>traditional calcium 2.5 g/d</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium Pidolate 0.405 g/d</td>
<td>+ 14,5 %</td>
<td></td>
</tr>
<tr>
<td>traditional calcium 2.5 g/d</td>
<td>+ 7,9 %</td>
<td></td>
</tr>
</tbody>
</table>

- calciuria

<table>
<thead>
<tr>
<th></th>
<th>Calcium Pidolate 0.405 g/d</th>
<th>traditional calcium 2.5 g/d</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium Pidolate 0.405 g/d</td>
<td>- 7,6 %</td>
<td></td>
</tr>
<tr>
<td>traditional calcium 2.5 g/d</td>
<td>+ 17,9 %</td>
<td></td>
</tr>
</tbody>
</table>

- hydroxyprolinuria

<table>
<thead>
<tr>
<th></th>
<th>Calcium Pidolate 0.405 g/d</th>
<th>traditional calcium 2.5 g/d</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calcium Pidolate 0.405 g/d</td>
<td>- 23,8 %</td>
<td></td>
</tr>
<tr>
<td>traditional calcium 2.5 g/d</td>
<td>- 11,2 %</td>
<td></td>
</tr>
</tbody>
</table>

The increase in calcaemia points to excellent gastrointestinal absorption. The reductions in calciuria and hydroxyprolinuria, greater than those with traditional calcium therapy, demonstrate the degree of calcium binding and particularly the incorporation of the pyrrolidine carboxylic ion in the organic bone framework.

CALCIUM PIDOLATE : ENCOURAGING RESULTS

Whatever the age

Percentages of positive results:

From 3 months to 5 years (n=35)

97 %

From 15 to 30 years (n=56)

64 %

From 31 to 55 years (n=26)

62 %

From 56 to 80 years and over (n=50)

67 %

BENEFITS OF CALCIUM PIDOLATE
Ease of use

As an active substance Calcium Pidolate offers numerous technical benefits. A white, odourless crystalline powder, Calcium Pidolate is extremely soluble and easy to process regardless of the manufacturing conditions of pH, temperature, humidity, etc.

Possible dosage forms

- ampoules for drinking
- ampoules for injection
- gel-sachet
- gel-vials
- tablets
- effervescent tablets
- powder sachets
- effervescent powder sachets
- syrup

Safety

These different dosage forms are always remarkably well accepted by all ages, both by children and the elderly.
The results from the following clinical trials relate to almost 500 cases. A minor digestive problem was reported in less than 1% of cases. Numerous authors have stressed the excellent tolerability of Calcium Pidolate. Others do not even bother to mention tolerability in their studies as, based on their experience, they tend to take it for granted.

BENEFITS OF CALCIUM PIDOLATE

SUMMARY OF LABORATORY STUDIES

PHYSICAL CHEMISTRY

Calcium Pidolate, or calcium pyrrolidone carboxylate (C$_{10}$H$_{12}$O$_6$N$_2$), is a white, amorphous odourless powder with a slightly salty and bitter taste. It is very soluble in water. In a 10% aqueous solution, its pH is between 5 and 7.

Physico-chemical measurement

Various comprehensive studies have been conducted to obtain a more accurate understanding of the physico-chemical behaviour of Calcium Pidolate. Accordingly, the following were determined in relation to other calcium salts and organic acids:

- the dissociation constants $K_1$
- the calcium association constant $C_1$

The relationship between these two constants was calculated. The resulting $K_1/C_1$ is some 30 to 50 times higher for Calcium Pidolate than for the other reference acids (calcium lactate, calcium gluconate). Its innovative physico-chemical characteristics mean that Calcium Pidolate possesses particularly favourable metabolic properties as regards gastrointestinal absorption, circulatory transport and protein binding.

TOXICOLOGY
**Acute toxicity**

- i.v. route LD$_{50}$: rabbit: 780 mg/kg, mouse: 357 mg/kg
- i.p. route LD$_{50}$: rat: 333 mg/kg
- oral route: the LD$_{50}$ has been investigated by two different authors (5,12) in four animal species: mouse, rat, rabbit and dog.

*No mortality was observed at the maximum physically administrable doses in the various animal species (from 6 to 10 g/kg Calcium Pidolate).*

**Subacute toxicity**

- i.v. route: rabbit: no mortality at the dose of 34 mg/kg/day

**Chronic toxicity**

- oral route: rat: 1 g/kg/day for 100 days. No mortality. No disruption of growth or behaviour. No macroscopic or microscopic lesion of the main viscera.
- oral route: rabbit: 500 mg and 1 g/kg/day for 16 weeks. No mortality. No impairment of growth, behaviour or laboratory values. Normal histological results.
- oral route: beagle dog: 400 and 800 mg/kg/day for 160 days. No mortality. No disruption of growth, behaviour or laboratory values. Normal histological results.

**Teratogenicity**

Study in two animal species: Wistar rat and New Zealand rabbit receiving 1 g/kg/day Calcium Pidolate throughout pregnancy. The parameters observed were the number of live foetuses, the number of stillborn foetuses, average litter weight, malformations and any resorptions.

*No significant difference from the control animals was observed.*

**PHARMACOKINETICS**

**Absorption**

Absorption of 100 mg/kg Calcium Pidolate by rats was followed by increases in serum calcium levels of 65 % in 40 minutes and of 100 % in 3 hours. In humans, the administration of 0.405 g orally resulted in a highly significant (p<0.001) increase in serum calcium levels of +14.5 %, whereas a reference calcium, administered at a dose of 2.5 g, i.e. 6 times higher, only produced a non-significant increase in calcaemia of 7.9 %.

*The gastrointestinal absorption of Calcium Pidolate is clearly superior to that of the other calcium therapies currently employed.*
Pharmacokinetics

Some very interesting pharmacokinetic results were obtained by L. ROBERT et al. (32) thanks to the use of labelled Calcium Pidolate molecules. Two different isotopes were used and these were detected, and their emissive activity measured, using a scintillation counter. The results were expressed in "counts per minute" (cpm) in the different organs.

Double radiolabelling was performed:

- firstly with carbon 14 for labelling the pyrrolidine carboxylic ion,
- secondly with calcium 45 to plot the fate of the mineral ion.

These studies demonstrated:

- that calcium is preferentially taken up by the skull and long bones (60 to 100 times greater than in other tissues).
- that the pidolate ion is also distributed throughout the skeleton, but in smaller quantities, and that a not insignificant proportion is taken up by other important organs: liver, spleen, brain, parathyroid, skin, etc.

It may be concluded that although the stronger specific emissive activity of Calcium Pidolate is observed in bone tissue, this molecule nevertheless plays a more general metabolic role in the body.

Metabolism

Column chromatography showed two optical density peaks corresponding to two radioactivity peaks:

- one corresponding to glycoproteins and the proteoglycans of the organic bony framework.
- the other to a metabolite of pyrrolidine carboxylic acid that recent research has shown to be important in the uptake of calcium by bone: gamma-carboxyglutamic acid.

In addition, electrophoresis of the hydrolyzing acids of organs shows that 85 % of the incorporated radioactivity corresponds to neutral amino acids, primarily proline and hydroxyproline. In metabolic terms, **Calcium Pidolate is involved in the incorporation in bone of proline and hydroxyproline, the biosynthesis of non-collagenous proteins of the bone: sialoglucoproteins, "Ca binding proteins", proteoglycans that play an important role in the bone uptake of calcium.**

PHARMACOLOGY

Recalcifying activity

Numerous pharmacological studies have investigated the recalcifying activity of Calcium Pidolate, including the two already been mentioned that investigated:

- the recalcification of an experimental matrix,
- and humoral recalcification.

Others worth mentioning include:

- A study on recalcification by Calcium Pidolate of a bone powder from which the calcium has been extracted using either 20 % EDTA or 0.6 N hydrochloric acid.
- A study on the recalcification of the rat skeleton receiving a calcium-free diet for 90 days. The degree of demineralization was assessed by means of x-rays and histological examination. The skeletons of rats treated with Calcium Pidolate showed almost normal opacity.
Experimental anti-osteoporotic activity

This study has also been mentioned above. An osteoprotic index was calculated for each treated group based on the scores obtained during the various radiographic investigations. *This index was less adversely affected in the animals treated with Calcium Pidolate.*

Stimulation of bone anabolism

*Labelled molecules* were also used in this study: *carbon 14* for the investigation of proline and, derived from this, the biosynthesis of hydroxyproline; and *sulfate S 35* for the investigation of the glycosaminoglycans. The results demonstrated:

- an increase in the binding of proline and the biosynthesis of hydroxyproline in the group of animals treated with Calcium Pidolate,
- stimulation of the biosynthetic activity of the glycosaminoglycans (increased incorporation of S 35) in the same group,
- the existence of a clear dose-response relationship in this biosynthetic effect of Calcium Pidolate.

References:

12. Gazave, J.M. Expertise toxicologique et pharmacologique du soluté buvable de pyrrolidone Carboxylate de Calcium (non publiée).
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