FERRIMANNITOL OVALBUMIN (FMOA)

PRODUCT PROFILE
Brief Introduction


Ferric Albuminate (egg ´s) is also described in:

- Index Merck XI Ed. Pages 3953-3958 (Ferric Albuminate).
- European Core Inventory (ECOIN) Unit 960 column 2 page 480.

Chemical Structure

- Molecular Formula 
  \((\text{Fe}_2\text{O}_3)_{242}(\text{C}_6\text{H}_{14}\text{O}_6)_{208}(\text{Protein})_2\)
- Apparent molecular weight 200 kD
- Proteic Fe = 14 - 18 %
- Proteic Nitrogen 7,5 - 9,0%

Production Method

Complex of ternary character formation through reaction among \(\text{Fe}^{3+}\), mannitol and properly hydrolyzed ovalbumin.

Pharmacological Profile

It can be concluded through studies carried out at Syntex S.A. that the product is an effective anti-anemic agent and that it presents a marked absence of gastric irritation in wide contrast to recognized products such as ferrous sulfate.

Another important datum to be considered is that this new therapeutic availability is totally alien to “slow” kind viral contaminations, in that it contains ovalbumin as proteic basis instead of mammal origin proteic derivatives such as ferritine. Apart from that, production method is adequately validated for the elimination of conventional viruses.

Studies carried out at Syntex S.A. include:

- Induced anemia assay. Ferrous Sulfate and Ferrimannitol Ovalbumin effects comparison.
- Gastric tolerance. Ferrous Sulfate and Ferrimannitol Ovalbumin comparative test.
Clinic Profile

Syntex has not carried out assays of dose fixation for humans, although product´s clinical experience in pharmaceutical market allows us to conclude that the dose is 80 mg Fe per day.

Presentation

- Containers of freeze dried powder or Spray-dried powder.
- Hydroglyceric solution of 20 mg Fe/g.
INDUCED ANEMIA ASSAY

FERRIMANNITOL OVALBUMIN (FMOA) AND FERROUS SULFATE (FS)

EFFECT COMPARISON
The aim of this test was to find out the capacity of Ferrimannitol Ovalbumin (FMOA) through oral via to correct induced iron deficiency anemia in rats. Its effect was compared to Ferrous Sulfate (FS), the most used drug to correct iron deficiencies.

Animals: 30 Wistar male rats weighing 170-230gr were used. From day 0 and during the entire assay they were fed with whole cow milk only. Animals were accommodated in plastic cages with shaving as bed.

Hematologic Techniques

Hemoglobin concentration was determined as cyano hemoglobin with a kit to that end (Hemoglowiener, Wiener Lab., Rosario), and the hematocrit was determined in a centrifuge for micro hematocrit. Heparin was used as anti clotting.

Procedure

On day 0, 2.5 ml blood was extracted through peri-orbital puncture from each rat, using light ether anesthesia. This procedure was repeated at regular intervals until the media hemoglobin concentration decreased to 50%. At that moment, 3 (three) groups of 8 (eight) rats each were formed, and they were fed exclusively with milk. Each group was daily administered oral via with the following products:

Group 1: distilled water 0.5 ml/100gr.
Group 2: SO$_4$ Fe in distilled water 0.5 ml/100gr.
Group 3: FMOA in distilled water 0.5 ml/100gr.

Groups 2 and 3 iron dose was 2 mg/kg. Administration was done using a syringe and a plastic cannula via esophagus. Solutions were prepared fresh and right before administration.

On day 15, after treatment was started, blood was extracted, hemoglobin and hematocrit were determined and the assay was considered to be finished.

Results and comments

Blood was extracted from animals on days 0; 8; 14; 21; 34 and 40. In table 1, hematologic variables and corporal weight evolution can be seen during that lapse.

Hemoglobin concentration value slowly decreased at the beginning and then faster as time elapsed.

Administration of 2mg iron be FS or FMOA once a day for 15 days, reverted the tendency to reduce hemoglobin and hematocrit while non treated animals did not (Table 2).
These results indicate that FMOA in these experimental conditions and doses can correct hematological variables modified by iron deficiency and/or loss.

Pharmacologic iron administration is normally done with ferric derivatives, such as sulfate, because they are salts that are absorbed far better than ferric ones. These, unlike the former, produce precipitates that are not soluble to intestinal pH, making absorption almost null.

Though, ferrous salts are inconveniently irritating to digestive mucosa. On the other hand, it is well known that the iron absorbed in the diet is of ferric type. For this reason, it was thought that a trivalent ferric composition with good intestinal absorption and non mucosa irritation effect could be developed. FMOA proved through this assay to be a good candidate to comply with these requisites given that in previous assays it showed little or null irritating capacity over digestive mucosa.

**TABLE 1**

Corporal weight evolution, hemoglobin and hematocrit concentration during anemia induction.

<table>
<thead>
<tr>
<th></th>
<th>Day 0</th>
<th>Day 8</th>
<th>Day 14</th>
<th>Day 21</th>
<th>Day 34</th>
<th>Day 40</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Corporal weight (g)</strong></td>
<td>210 (27)</td>
<td>209 (23)</td>
<td>225 (41)</td>
<td>229 (60)</td>
<td>--</td>
<td>--</td>
</tr>
<tr>
<td><strong>Hemoglobin (g/100 ml)</strong></td>
<td>15.7 (0.9)</td>
<td>13.2 (1.0)</td>
<td>13.3 (1.1)</td>
<td>11.6 (1.5)</td>
<td>9.8 (1.1)</td>
<td>7.8 (1.3)</td>
</tr>
<tr>
<td><strong>Hematocrit</strong></td>
<td>49 (2)</td>
<td>45 (3)</td>
<td>43 (3)</td>
<td>--</td>
<td>--</td>
<td>31</td>
</tr>
</tbody>
</table>

* Media value (Standard deviation)

**TABLE 2**

Data obtained after 15 days of iron derivatives treatment: Corporal weight, hemoglobin and hematocrit concentration in rats suffering from induced iron deficiency anemia

<table>
<thead>
<tr>
<th></th>
<th>Control Rats</th>
<th>Rats under SO₄ Fe treatment</th>
<th>Rats under FMOA treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Corporal weight (g)</strong></td>
<td>248 (27) (a)</td>
<td>251 (28)</td>
<td>251 (40)</td>
</tr>
<tr>
<td><strong>Hemoglobin Concentration</strong></td>
<td>6.5 (1.1)</td>
<td>10.2 (1.1) *</td>
<td>10.6 (1.0) *</td>
</tr>
<tr>
<td><strong>Hematocrit</strong></td>
<td>32 (4)</td>
<td>45 (3) *</td>
<td>45 (5) *</td>
</tr>
</tbody>
</table>

(a) Media Value (Standard deviation)
(*) Presenting significant differences with controls (Newman Keuls Test) p<0.05.
GASTRIC TOLERANCE
COMPARATIVE ASSAYS
BETWEEN
FERRIMANNITOL OVALBUMIN
AND
FERROUS SULFATE
Materials and methods

Twenty Wistar male rats weighing 300/400 gr were used. Animals were accommodated in individual plastic cages with shaving as absorbent bed. They were divided into three groups: a control group of 6 (six) animals and two groups of 7 (seven) animals each (SO$_4$Fe and FMOA).

Pilot test was done as follows:

**Day 1:** Fast started at 5 (five) in the afternoon. During the test, the animals were fed with sucrose solution 0.5% in a water top ad limitum and nothing else during the test.

**Day 2:** At 4 (four) in the afternoon, treatments were administered orally.

**Day 3:** Ditto day 2.

**Day 4:** At 9 (nine) in the morning, the treatment was administered. At 2 (two) in the afternoon, animals were sacrificed by asphyxia with ethyl ether, stomachs were extracted and examined; duodenum ‘s first part was taken out and examined after a longitudinal cut.

   - **Group 1:** Fed with a top of water.
   - **Group 2:** Ferrous Sulfate.
   - **Group 3:** Ferrimannitol Ovalbumin.

Dose for both groups were equivalent to 200 mg/kg of dissolved and elemental iron facilitating administration of 0.5 ml/100 gr of corporal weight this way. The drugs used were SO$_4$Fe7H$_2$O and lyophilized FMOA.

Results

**Group 1 (Control)**

None of the animals presented gastric dilatation. In some individuals a slight irritation process extended to the duodenum could be observed. None died.

**Group 2 (Ferrous Sulfate)**

All animals presented an intense dilatation along the stomach, especially in fundic gland area as the most important hit. Important iron impregnations in the mucosa were observed, forming thick conglomerates in some cases.

All animals showed clear signs of gastritis to different degrees. This kind of irritation was also observed in the duodenum. One animal died during the tests.
Group 3 (Ferrimannitol Ovalbumin)

None of the animals showed gastric dilatation. An irritation slightly higher than control animals though similar to it was observed in general. Two individuals showed some FMOA precipitations apart from small swellings.

Discussion

The slight irritation observed in individuals, was caused doubtless by the long fast that the animals were subjected to. Thus, during that time the gastric juice was in an unprotected organ due to lack of food. The passage of gastric juice through the intestines could explain the irritation observed in the higher part of the organ.

This experimental condition allows a clearer observation of irritating effects of the orally administered compound.

The magnitude of the dose administered and the fact of being a sensitized organ, proposes a limit situation far more severe than what was considered in practice.

Considering all aforementioned and from the comparative analysis of both treatments, it can be assured that FMOA is much less aggressive for the digestive mucosa than Ferrous Sulfate (SO$_4$Fe).

The Ferrous Sulfate irritating nature has been previously described in different situations and circumstances, clinically and experimentally.

Taking into account the magnitude of the dose administered, it shall be correct to establish that in normal conditions, with a lower dose -therapeutic- and in presence of food in the digestive tract, the FMOA would not be irritating to gastrointestinal mucosa.
Control Group

Stomachs from two rats having pale reddish coloured mucosa.
Group 2 (FS)

Rats stomachs treated with Ferrous Sulfate:

Both show Ferrous Sulfate deposits. An intense mucosa irritation can be also observed. In left sample the irritation is more intense while the right sample shows a ferrous deposit in the duodenum.
Group 3

Stomachs of two rats treated with FMOA. The irritation is similar to control samples. Upon right side, some FMOA deposits can be observed. Duodenum irritation is almost the same as control ones.
Typical stomachs appearance before they are opened for assessment. The smaller one belongs to a rat treated with FMOA while the bigger and darker belongs to a rat treated with Ferrous Sulfate.