



NEONATAL SCREENING FOR CLASSIC GALACTOSEMIA. SYSTEMATIC REVIEW.

SUMMARY

Introduction: Galactosaemia is an autosomal recessive disease characterised by incapacity to metabolise galactose in glucose. In classical galactosaemia there is a galactose-1-phosphate uridylyltransferase (GALT) gene deficiency and galactose-1-phosphate cannot be converted to glucose 1-phosphate. Consequently, in the case of intake of lactose (which is hydrolysed in the intestine to glucose + galactose), whose main source in the human is milk and dairy products, galactose and galactose 1-phosphate accumulate in blood and tissue. Neonates who present galactosaemia tend to show feeding difficulty and general toxic manifestations during the first weeks of life, including vomiting and diarrhoea, weight loss, jaundice, hepatomegaly and ascites. If lactose is not withdrawn from the diet, the toxic syndrome may progress during the first weeks and give rise to more severe complications. The progressive accumulation of galactitol in the crystalline lens, a metabolite of galactose, can result in the denaturation and precipitation of lenticular protein and formation of cataracts. Accumulation at the level of other organs such as the brain, liver and kidney can lead to poor renal tubular reabsorption, cirrhosis, liver failure or cerebral oedema. Galactosaemia can cause fulminant sepsis due to *E. Coli* during the first weeks of life. The aim of neonatal galactosaemia screening is a presymptomatic identification and early treatment of galactosaemia, in order to reduce morbidity-mortality and possible disease-related disabilities.

Objectives: This assessment report was drawn up at the request of the National Health System Interterritorial Council's Services, Insurance & Finance Committee, in response to a proposal from the Galician Regional Health Authority. Its overall objective was to analyse existing evidence on neonatal galactosaemia screening to help decide on its introduction into the National Health System service portfolio. A specific objective was to assess different aspects of the disease, treatment and screening test which might serve to support decision-making. Secondary objectives were to recover information on the different screening strategies used world-wide (diagnostic tests, confirmatory tests, cut points).

Methods: In order to ascertain if the technology meets the requirements for implementation of national screening in Spain set out in the "Population Screening Framework Document", we carried out two systematic searches of the scientific literature in the leading biomedical databases. The first search was conducted in February 2014, and was targeted at searching for information about internationally implemented screening programmes (detection rate, sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV)). The second search was conducted in May 2014, and focused on retrieving all the relevant information about the disease (definition of galactosaemia, clinical characteristics, morbidity-mortality, quality of life, treatment). Papers were selected by two independent reviewers in accordance with a set of pre-defined inclusion/exclusion criteria, and the quality of the scientific evidence was assessed based on the "Oxford Centre for Evidence-Based Medicine 2011 Levels of Evidence" scale.

Results and discussion: Based on three retrospective studies which reported information on clinically diagnosed cases, it can be anticipated that most neonates with classical galactosaemia will develop characteristic symptomatology of the disease within the first 2 weeks of life and most of the severe cases and deaths due to septicaemia will occur in this period. According to two descriptive studies, the mean age of hospitalisation for severe symptomatology is 12-13 days. The available evidence, all deriving from surveys, cross-sectional studies or retrospective case series with important methodological limitations (Oxford Centre for Evidence-Based Medicine level-4 evidence) shows that, despite galactose restriction since birth, classical galactosaemia can lead to important complications developing in the medium-long term. Patients usually present reduced intelligence coefficients $\leq 80-85$



(32%-83%), speech/language disorders (22%-91%) and motor disorders (66%). In women, galactosaemia is associated with hypergonadotropic hypogonadism (90%-100%). According to two of the studies identified, galactosaemia negatively affected patients' quality of life.

Early lactose restriction is thought to succeed in preventing the most acute complications in the neonatal period. In general, consensus documents recommend that, in the case of diagnosis or clinical suspicion of galactosaemia, breast milk and lactose formulae should be withdrawn and replaced by lactose-free formulae –restricting the diet to soy-based proteins- for life. There is no agreement regarding the exclusion of fruit, green leafy vegetables and other products not having high galactose content, and specific dietary recommendations tend to vary from country to country. A recent study reports a similar result for strict compliers and non-compliers with the diet, and a few authors express the need for additional studies to assess the possibility of galactose intake liberalisation after one year, with strict surveillance. At present, there is likewise no consensus on the treatment of Duarte variants or mild heterozygous forms, though recent studies also suggest that dietary restriction might not be justified in these patients. All the studies retrieved were retrospective and lacked statistical power for drawing up guidelines (Oxford Centre for Evidence-Based Medicine level-4 evidence).

From the few documents retrieved by the search, it can be concluded that currently there is no standard protocol for newborn screening of classical galactosaemia. In most of the protocols, the initial test was based on total galactose determination (galactose and erythrocyte galactose 1-phosphate), and determination of the GALT enzyme was used either as a second-tier or confirmatory test, though in some countries such as Sweden screening begins with semiquantitative determination of the GALT enzyme. Protocols generally differ considerably in terms of strategy, analytical methods and cut points. Galactose determination is usually performed using qualitative (Guthrie test) or semiquantitative microbiological methods (Paigen), microchemical tests (Fujimora, rapid GAL-DH test), and colorimetric or tandem mass spectrometry methods. Among the most widely used methods for GALT determination are the Beutler test and the radioactive assays. Many variants of all these procedures have been described, and currently there are many simple kits for same-day testing. A factor that all screening procedures have in common is that they are performed on the basis of a neonate heel-puncture blood sample routinely collected for the screening of other congenital metabolic diseases.

Based on data drawn from four descriptive studies, it is estimated that all programmes have a sensitivity of 100% and a specificity of 99.9%, though these values should be interpreted with caution since none of the studies have confirmed negative cases. The screening protocols used in the various countries are based on different screening tests, cut points and confirmatory tests, and thus differ with regards to the definition of a positive case. In the five publications included in the current review, the false-positive rate ranged from 0.0005% to 0.25%. The PPV ranged from 0% to 64.3%: the only studies that obtained a PPV of over 20% were those that reported Swedish and Galician Regional screening programme data. The Swedish programme uses a two-tier strategy (the Beutler test with the Rapid GAL-DH test). In the Galician Autonomous Region, the screening test comprises four steps, determination of galactose 1-P by tandem mass spectrometry (MS/MS), parallel determination of reducing substances in urine, thin-layer chromatography and high performance thin layer chromatography. In the calculation of the PPV, the recall (follow-up testing) rate for the second sample was not taken into account because there was no information on this aspect. The PPV was very low in the Danish screening programme, which relies exclusively on MS/MS as its diagnostic test.

There are insufficient data to clarify to what extent screening is really effective in reducing the appearance of severe adverse effects and/or neonate mortality. The screening studies retrieved which reported clinical data display great variability with regard to the presentation of symptomatology until diagnosis and mortality (0%-100%). In the case of Sweden, only 4 patients with galactosaemia were



reported to have survived before implementation of screening in 1967 and only 1 was reported to have died since 1967: in this country, diagnosis is made during the first week of life.

Currently, there are no adequate comparative studies that can allow for determining the effectiveness of neonatal galactosaemia screening against implementation of other measures designed to prevent severe acute complications (surveillance programmes, opportunistic screening). The only comparative data comes from the United Kingdom (UK) paediatric surveillance programme survey, which points to similar incidence of severe cases and mortality in the regions which have and have not implemented a galactosaemia screening programmes, but does not take into account the characteristics of public health-care planning in the UK.

Conclusions:

- As classical galactosaemia progresses, severe neonatal complications can develop. With early detection and treatment, it is estimated that most of the severe acute complications and deaths due to septicaemia which occur in the first weeks of life could be prevented.
- Existing information highlights that fact that, despite early dietary treatment, patients with classical galactosaemia may present with important complications in the medium-long term (cognitive deficiency, speech/language disorders, motor disorders, etc.).
- Based on data drawn from four descriptive studies, the screening programmes would be estimated to have a sensitivity and specificity of around 100%, though these data should be interpreted with caution since there are no studies that confirm negative cases.
- Screening tests generally display a high FP rate and a low PPV. The only studies that obtained a PPV of over 20% were those that reported data from the Swedish screening programme and the Galician Regional programme, which uses four steps.
- Screening protocols differ considerably in terms of screening strategy, analytical methods and cut points (GALT activity $\leq 15\%$ to $\leq 30\%$). Depending on the cut point, there is a different probability of having classified mild heterozygous forms or benign Duarte variants as positive. In the long term, the lack of differentiation of certain mild heterozygous forms and the Duarte variant could be regarded as an adverse screening effect.
- There is not enough information to establish whether screening is really effective in reducing the frequency of severe adverse effects or neonate mortality. On the basis of indirect assumptions and descriptive data from the Swedish and West German programmes, screening could be assumed to be capable of reducing the risk of mortality/morbidity provided that the screening results are obtained during the first week of life, though this assumption should be confirmed by suitably designed studies.
- Currently, there are no comparative studies that would make it possible to establish whether neonatal galactosaemia screening is really superior to other secondary prevention measures aimed at preventing severe acute complications and mortality in the neonatal period (alert protocols, opportunistic screening).
- The programme's cost-effectiveness has still to be assessed and its potential impact on the national health system is unknown.



Recommendations:

- If the implementation of a national galactosaemia programme were to be considered, it would be important to reach a consensus on the screening protocol to be used, based on the results of the scientific evidence and expert opinion.
- To ensure the benefit of screening, the time interval between taking the sample and obtaining the results must be minimised.
- Information systems must be maintained to monitor the benefits/risks of screening.

