Hypospadias and Environment : a Case-Control Study

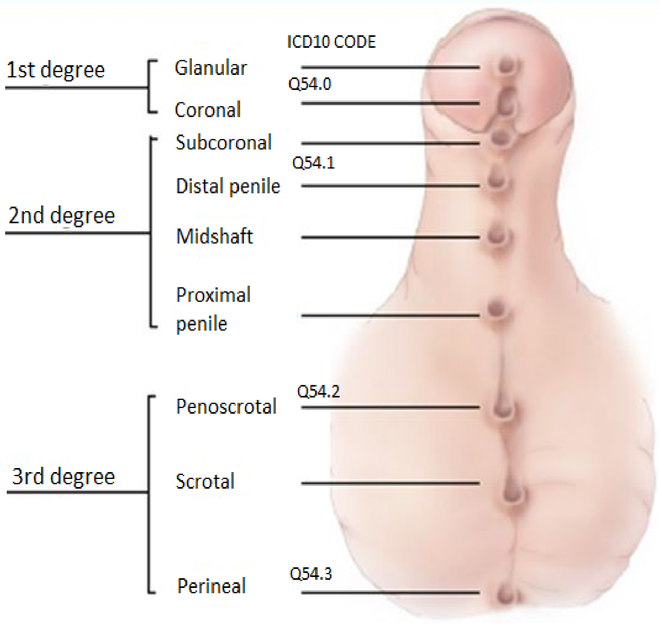
Lucas Michon*, Elisabeth Gnansia*, Ludivine de Brosses*, Sourour Addad*, Catherine Cuoq*, Véronique Dimper*, Emmanuelle Amar*

*Registre des Malformations en Rhône-Alpes, REMERA

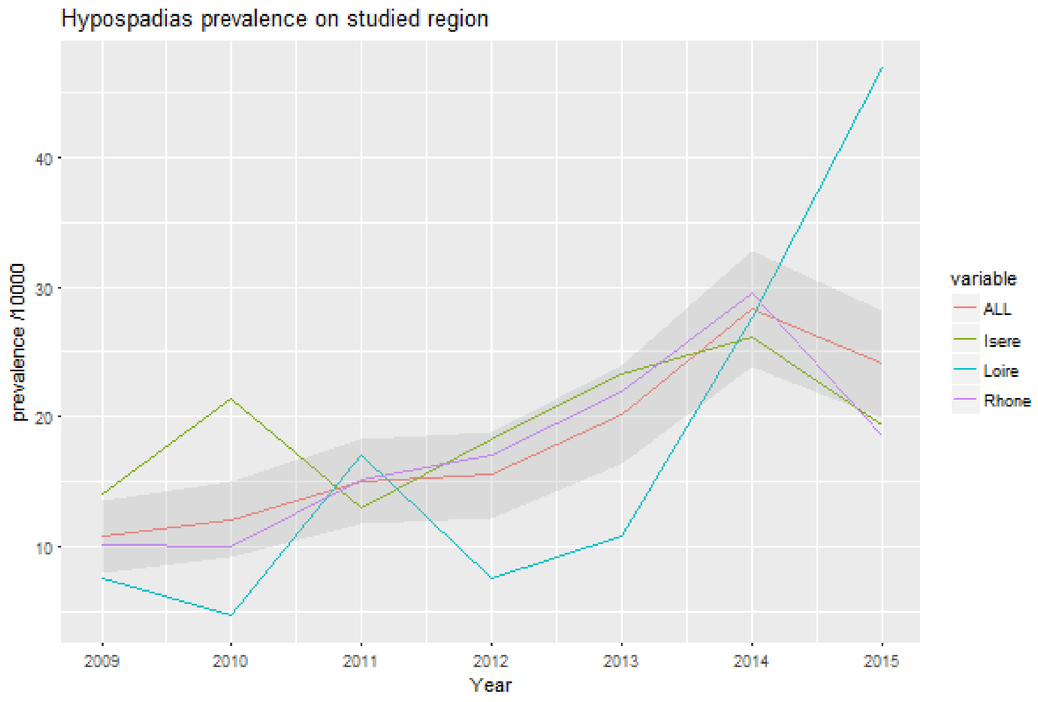
lucas.michon@remera.fr

www.remera.fr

Introduction



Hypospadias is a genital malformation characterized by an ectopic position of urethral meatus, with different degrees in function of this position. Monitoring carried out by Remera (Registre des Malformations en Rhône-Alpes) shows a significantly increasing prevalence of hypospadias since 2009. As Endocrine Disruptors (ED) definition and their consequences on fetal development are a debated issue, evaluating ED exposure during the first month of pregnancy could contribute to the comprehension of observed increase of hypospadias. Therefore we analyzed the registry data to determine whether factors such as pesticide exposure were involved, using multivariate statistical methods, machine learning, and spatial data mining.



Materials and methods

Inclusion criteria. Cases and controls were identified in the registry. Cases (n=479) were male children born in Rhône, Isère, or Loire between 2009 and 2015 with an isolated hypospadias (ICD10 code Q54). Controls (n=1142) were male children born in the same period and region, with a unique and isolated malformation. Malformations were selected as controls if and only if no correlation with endocrine disruptors exposure is known and if they are independent from genital abnormalities.
Explanatory variables.

- Utilised Agricultural Area (UAA) rate in maternal municipality
- Maternal 'Département' of origin
- Mother's and father's age and mother's BMI, signs of subfertility
- Birth year and conception month
- Agricultural type in maternal municipality (wine and fruit agriculture)
- Gestational age (weeks after LMP)
- Parity
- Assisted reproductive technology ART

Statistical Analysis. We used univariate logistic regression for variables selection (threshold : p<0.2) then multivariate regression with backward exclusion procedure according to Akaike Information Criterion. Hosmer-Lemeshow test and cross-validation were performed to validate the model. We introduced a statistic to test the homogeneity of the prevalence (H0) upon the region. If H0 was rejected, it tends to demonstrate involvement of environmental factors and then we performed *post hoc* spatial interpolation (k nearest neighbors, Inverse Distance Weighting and Voronoï diagram) and data mining algorithms to found high risk zones. Algorithms are based on Kulldorf statistic with multiple testing correction or on a hierarchical approach, with stop criterion when contiguous regions are too different. We further note p-value p (significant threshold for $p < 0.05$), adjusted odds-ratio aOR and 95% confidence interval CI.

References

[1] Carissa M. Rocheleau, Paul A. Romitti, and Leslie K. Dennis. Pesticides and hypospadias: A meta-analysis. *Journal of Pediatric Urology*, 5(1):17 – 24, 2009.

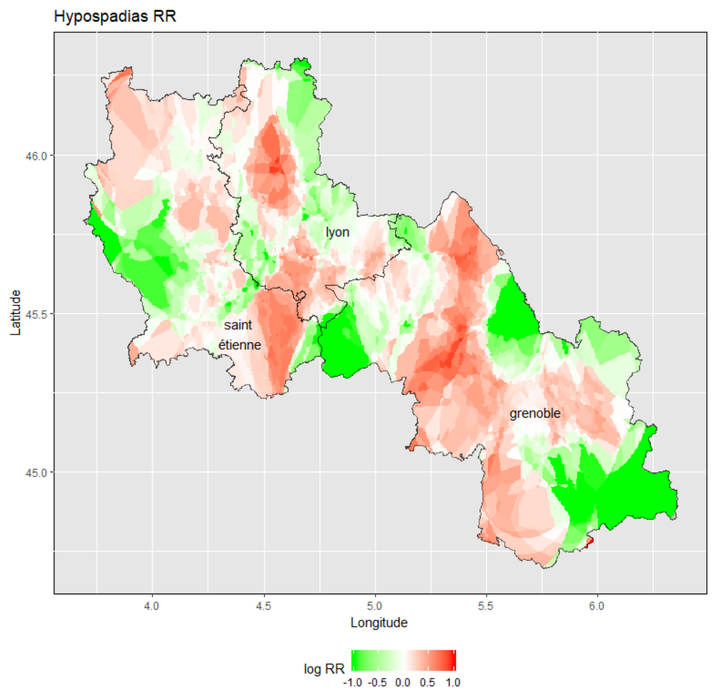
[2] Laura Gaspari, Françoise Paris, Claire Jandel, Nicolas Kalfa, Mattea Orsini, Jean Pierre Daurès, and Charles Sultan. Prenatal environmental risk factors for genital malformations in a population of 1442 french male newborns: a nested case–control study. *Human Reproduction*, 26(11):3155–3162, 2011.

[3] Elodie Haraux, Pierre Tourneux, Christelle Kouakam, Erwan Stephan-Blanchard, Bernard Boudailliez, Andre Leke, Celine Klein, and Karen Chardon. Isolated hypospadias: The impact of prenatal exposure to pesticides, as determined by meconium analysis. *Environment International*, 119:20 – 25, 2018.

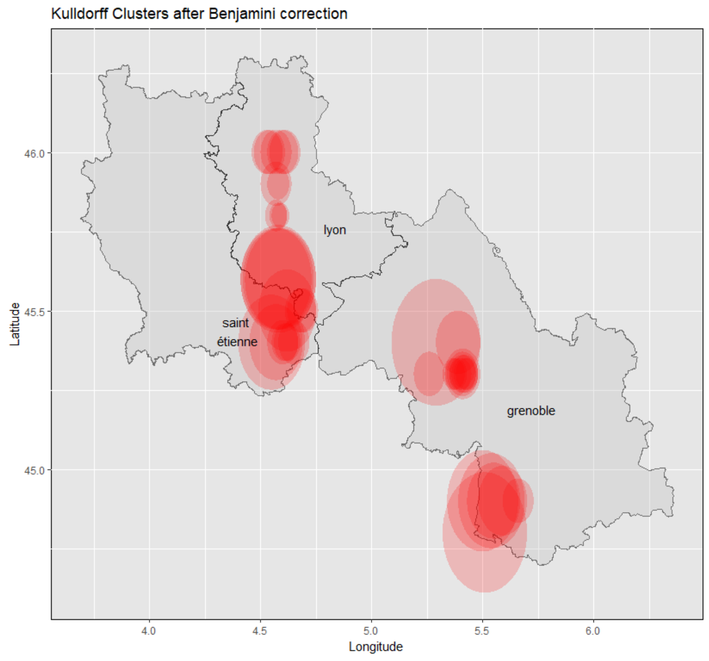
Results

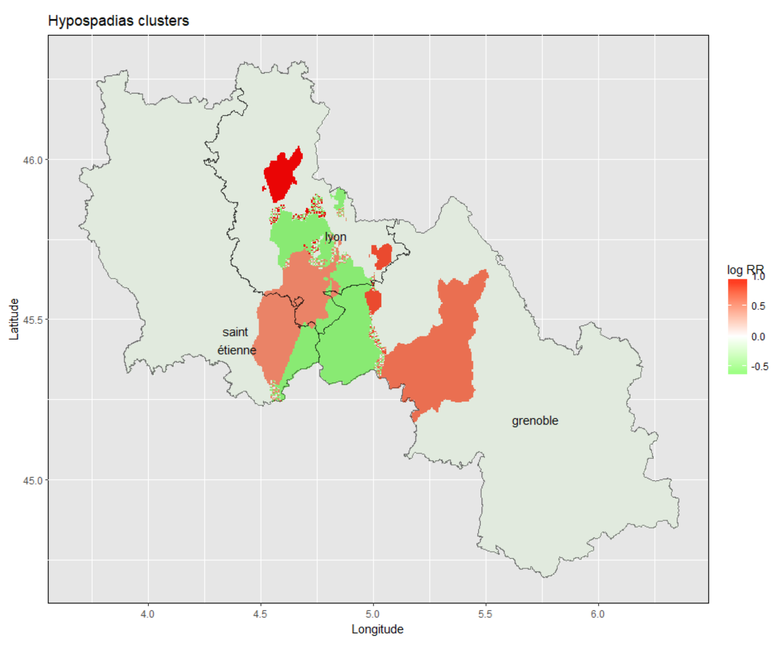
	aOR	p	adjusted CI
UAA rate >40%	1.42	0.02	[1.06 ; 1.90]
Birth year	1.09	0.005	[1.03 ; 1.16]
May to October conception	1.35	0.01	[1.07 ; 1.71]
Term <32 weeks after LMP	2.40	0.002	[1.36 ; 4.19]
Parity	0.79	0.0002	[0.70 ; 0.90]

Mothers living in rural areas had a significant increased risk to bear a boy with hypospadias. These mothers are potentially more exposed to pesticides. Moreover, we observed a periodicity with high risk season from May to October. We chose a priori this season because atmospheric analysis showed that there is the highest pesticides concentration in air and it matched with spreading periods. We exclude hypothetic periodic confusion factors like diseases. Moreover, we observed a decreasing risk with parity, *i.e.* each boy had a lower risk than his elder brother. As ART and parent's age do not appear as risk factors, we tend to **reject subfertility as an explanation** for it. An hypothesis is that some **pollutants with high bioaccumation capacity could be a risk factor**. Indeed, it was proven that concentration of those molecules (*e.g.*, PCB or dioxin-like) in mother's body decreases after each breastfeeding. Finally, severe prematurity, sign of ischemia, appeared as risk factors and birth year too, after controlling potential confusion bias. Proximity to fruit or wine agriculture did not appear as risk factor.



We rejected H0, homogeneity of prevalence ($p < 10^{-10}$). It tends to involve environmental factors in hypospadias development. We performed prevalence interpolation and spatial data mining algorithms. The results were matching. Three significant high risk zones were found whatever the method used. Beaujolais is the region with the highest relative risk. It is the most important wine zone in studied region (wine agriculture is known to be the most polluting) and the most polluted zone for several molecules (mancozeb, glyphosate, folpet, ...). The 'Bièvre' area, which is an agricultural region with a dominant of maize, wheat, and colza culture, emerged too. Like Beaujolais, this area is also affected with concerning levels of water pollution.





Conclusion

Our study tends to show the involvement of environmental factors in hypospadias development, which supports previous studies [1, 2, 3]. Indeed, higher hypospadias risk is correlated with a rural or highly polluted zones domiciliation and first months of pregnancy during the pesticides spreading seasons. Further studies with precise pesticide exposure quantification (*e.g.*, toxicological measurements) should be conducted to confirm the generated hypotheses.