Université catholique de Louvain
Faculté des sciences agronomiques
Laboratoire d'écologie des prairies

Restauration des couverts herbacés riches en espèces.

Frédéric JANSSENS

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Promoteur :

PEETERS A. (UCL)

Lecteurs :

Monsieur F. DEVILLERS
Monsieur J. DUFEY
Monsieur T. DUTOIT
Monsieur B. JEANGROS
Monsieur J. LAMBERT

Louvain-la-Neuve

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II. TABLE DES MATIÈRES

III. CADRE DE LA THÈSE

L'ambition de restaurer les couverts herbacés riches en espèces est relativement récente. La nécessité d'une action en faveur du maintien des milieux les plus intéressants était cependant déjà évoquée en 1912 à l'initiative de Jean Massart dans son livre "pour la protection de la Nature en Belgique". Une vingtaine d'années plus tard, les premières associations dont les actions visaient à mettre en réserve les sites les plus menacés voyaient le jour (Amis de la Fagne en 1935; Ardenne et Gaume en 1941, les Réserves Naturelles et Ornithologiques de Belgique en 1951,...). Le but de ces réserves, à l'instar de pièces de collection dans les musées, était de conserver la vie sauvage présente en évitant toute action susceptible de perturber la flore ou la faune.

A l'aube des années septante, un nouveau concept s'impose. Parent (1966) introduit, en effet, la notion de gestion. A partir de ce moment, la mise en réserve, passive, va être complétée par une gestion beaucoup plus active. Ce principe de gestion va peu à peu s'imposer dans tous les milieux et susciter de nombreuses publications scientifiques.

La légitimité de cette évolution doit, à ce stade, être discutée. En effet, en quoi la sauvegarde d'une flore diversifiée dans les couverts herbacés est-elle importante pour l'humanité ? Que perdrons-nous si cette diversité venait à disparaître ?

Le débat concernant la protection de la biodiversité à l'échelle de la planète a déjà fait couler beaucoup d'encre. Il n'est toutefois pas inutile de présenter succinctement quelques éléments qui ont nourri cette réflexion et qui s'appliquent directement au problème de la diversité botanique dans les couverts herbacés.

La problématique de la sauvegarde des prairies riches en espèces est assez particulière. En effet, le fait que ces prairies soient semi-naturelles, c'est-à-dire d'origine anthropique, est souvent un argument formulé contre leur sauvegarde. Il faut se rappeler que la végétation dans nos régions était, avant l'homme, essentiellement constituée de forêts. Il a fallu que celui-ci défriche et utilise le sol pour changer les choses. Les landes sont nées de ce défrichement. La civilisation évoluant, l'utilisation du sol s'est faite plus intensive, les sols ont notamment été enrichis, permettant la survie d'un nombre de plantes de plus en plus grand. C'est ainsi que sont nées les prairies semi-naturelles. Ces dernières décennies, l'intensification des pratiques agricoles s'est fortement accrue (par un emploi massif de fertilisants, par un changement des régimes de coupe, par des drainages, des semis,...). Le nombre d'espèces présentes dans ces prairies s'en est trouvé fortement amoindri. Les espèces les plus productives ont occupé l'espace au détriment des autres espèces pour en arriver finalement aux prairies comprenant moins de 10 espèces que nous connaissons actuellement. On peut donc raisonnablement se demander pourquoi protéger quelque chose qui n'existait pas naturellement, et que l'homme a finalement créé.

Les raisons pour lesquelles la biodiversité en général et la diversité végétale dans les couverts herbacés, en particulier, doivent être préservées se divisent classiquement en deux groupes : les raisons éthiques et les raisons anthropocentriques (Green, 1985).

Les raisons éthiques sont délicates à aborder car, relevant du domaine de la subjectivité, chacun les appréhende différemment. Beaucoup de gens considèrent la protection de la nature comme une question de conscience (Green, 1985; Norton, 1988).

D'un côté, certains voient dans l'être humain un être dominateur, disposant de la création comme bon lui semble (Genèse 1:26). Cette notion est loin de se cantonner aux traditions Judéo-Chrétiennes. Ces notions de supériorité humaine apparaissent dans bon nombre de religions. Ceci revient finalement à considérer l'homme comme un être "à part" de la nature, cette vision des rapports de l'homme avec la nature peut être qualifiée de "dualiste" (Maljean et al., 1997).

D'autre part, l'être humain est considéré comme une espèce vivant sur terre avec d'autres espèces dans un ensemble complexe auquel il appartient mais qu'il ne domine pas. Cette vision de la place de l'homme dans la nature est qualifiée cette fois de "systémique".

Comment ces courants de pensées peuvent-ils être interprétés dans le cadre de la préservation de la nature et, plus précisément, dans le contexte de ce travail, de la diversité botanique dans les couverts herbacés ?

La vision systémique amène à répondre par la négative à la question "a-t-on le droit de laisser ou de faire disparaître des espèces qui, comme nous, vivent sur terre ?". Ceci revient finalement à définir un droit à l'existence pour chacune des espèces vivantes (Ferry, 1992; Primack, 1993; Serres, 1990). La vision dualiste, par contre, ne verra une raison à la conservation de la nature que si l'homme peut en tirer un quelconque avantage (Primack, 1993). Ceci nous amène à la seconde catégorie de raisons justifiant la sauvegarde de la nature : les raisons anthropocentriques.

Les raisons anthropocentriques indirectes sont aussi assez difficiles à appréhender objectivement. La présence de prairies fleuries est considérée par beaucoup comme améliorant la valeur esthétique des paysages (Janssens et al., 1996). Une notion qui peut être liée à la valeur esthétique réside dans la perception culturelle de ces milieux. En effet, liés à un passé encore assez proche, ces couverts font partie d'un héritage auquel on peut être attaché sentimentalement. Cet héritage peut, bien entendu, être porteur de valeurs plus objectives dont on peut tirer profit.

Les raisons anthropocentriques directes sont liées aux bénéfices scientifiques et économiques d'une telle action. Une première raison concerne l'écosystème dans son ensemble. La perte d'une espèce peut paraître anecdotique à l'échelle de la biosphère. En réalité, les conséquences liées à la disparition d'une espèce végétale peuvent être d'une extrême importance. Cette disparition, par les relations étroites qui existent entre les individus d'un écosystème, peut entraîner l'extinction d'une autre espèce et, par une suite de mécanismes, déstabiliser l'écosystème dans son ensemble. Ehrlich (1988) estime que, si la perte de diversité continue pendant 100 ans au même rythme, les effets seront comparables à un hiver nucléaire...

La nature s'avère souvent être une mine d'or pour les chercheurs. Celle-ci a en effet, tout au long de son évolution, développé des mécanismes, des substances, des comportements qui ont permis aux éléments vivants qui en ont bénéficié de s'adapter à leur environnement. Il ne reste plus au scientifique qu'à reproduire et améliorer pour son propre besoin ce que la nature a déjà réalisé. Bon nombre de substances à vertu médicinale ou aromatique ont ainsi été découvertes (Primack, 1993; Nations, 1988, Norton, 1988). La diversité pouvant se définir à l'échelle des gènes, un intérêt scientifique à sa préservation peut aussi être trouvé à ce niveau. Par exemple, pour leur survie dans des conditions particulières, des plantes ont développé des gènes (de résistance à des maladies, au froid,...) directement utilisables en recherche.

Voilà présentés succinctement quelques éléments de réflexion à propos de la légitimité de la conservation de la nature. Ces éléments, et toutes les actions de sauvegarde qu'ils ont motivées ont mené le Conseil de l'Europe et ensuite les autorités belges à formuler les bases législatives de la conservation de la nature encore en vigueur à l'heure actuelle. C'est ainsi qu'est née, chez nous, la loi du 12 juillet 1973 sur la Conservation de la Nature.

Tout ceci n'a toutefois pas empêché une raréfaction du nombre et de la surface occupée par les couverts herbacés riches en espèces, menant finalement à une insularisation des quelques rares parcelles intéressantes. La notion de maillage écologique est née de ce problème. Il peut être défini comme un ensemble de biotopes reliés entre eux et permettant la conservation d'espèces sauvages sur un territoire local. Ce maillage est donc structuré autour de zones centrales (réserves naturelles,...) et constitué d'éléments linéaires (haies, berges de cours d'eau, talus,...) ou ponctuels (arbres, mares, petits prés fleuris,...). Cette notion a connu un succès fulgurant et oriente de nos jours beaucoup de prises de décisions, que ce soit au niveau national ou européen. C'est ainsi, par exemple, que le Comité Régional d'Organisation de l'Année Européenne de la Conservation de la Nature (1995) a sollicité les pouvoirs communaux afin qu'ils entreprennent des actions en faveur de la vie sauvage sur les bords de route. Dans la même année, les Plans Communaux de Développement de la Nature prenaient place chez nous. Ces PCDN n'ont d'autre objectif que de créer, à l'échelle communale, un réseau écologique. Sur la base d'un inventaire visant à estimer l'état de l'environnement présent et potentiel au sein de la commune, un programme d'actions est mis en place.

La mise en application d'un tel objectif n'est cependant pas aussi simple qu'il y paraît. La réalisation pratique de ce maillage écologique se heurte encore à beaucoup d'incertitudes. Parmi celles-ci, les conditions d'existence des couverts herbacés riches en espèces et les pratiques à adapter afin de les restaurer, sont loin d'être connues. C'est à ce niveau que s'insère l'objet de ce travail.

IV. OBJECTIFS

Le premier objectif de cette thèse concerne l'étude des relations entre la diversité botanique dans les couverts herbacés et les teneurs en éléments nutritifs du sol. Ces relations permettront de définir les teneurs en éléments nutritifs incompatibles avec l'existence des couverts herbacés riches en espèces.

Le second objectif, lié à la connaissance de ces teneurs, se rapporte à l'établissement d'une méthode rapide d'estimation de la potentialité d'un site à supporter un couvert herbacé diversifié.

Le troisième objectif s'applique aux sites présentant des teneurs en éléments nutritifs incompatibles avec l'existence de couverts herbacés riches en espèces. Diverses méthodes classiques ou originales visant à réduire ces teneurs seront explorées. L'objectif vise à proposer la méthode combinant facilité d'emploi et efficacité.

Le dernier objectif consiste en une étude de la germination d'espèces caractéristiques de ces milieux en fonction de ces teneurs en éléments nutritifs.

V. STRUCTURE DE LA THÈSE

A. CONSIDÉRATIONS GÉNÉRALES

Cette thèse est présentée sous forme d'une suite intégrée de cinq articles. Cette formule implique nécessairement des redites essentiellement au niveau des introductions et des matériels et méthodes. Cependant, l'avantage de ce choix réside dans l'indépendance de chaque article, et donc, dans la possibilité de lecture de chacune des parties sans devoir faire référence au reste du travail.

Les exigences éditoriales en matière de concision laissent peu de marge de manoeuvre pour une présentation ou une discussion générale du sujet. Celles-ci sont présentées dans la section suivante pour chacun des articles.

Suite à cette introduction, le travail se présente sous forme d'une série d'articles séparés, suivie des conclusions générales synthétisant les principaux résultats acquis.

Afin de faciliter la lecture de ce travail, les références bibliographiques sont rassemblées à la fin du manuscrit.

B. LISTE ET ARTICULATION DES ARTICLES

Les articles qui composent cette thèse ambitionneront tout d'abord la mise en évidence de certaines caractéristiques du sol susceptibles d'influencer l'existence de couverts herbacés riches en espèces. C'est dans ce cadre que des hautes teneurs en phosphore échangeable du sol se sont avérées fortement préjudiciables à la diversité botanique (articles 1 & 2).

1. *Janssens F., Peeters A., Tallowin J.R.B., Bakker J.P., Bekker R.M., Fillat F. & Oomes M.J.M. Accepté. Relationship between soil chemical factors and grassland diversity. Plant and Soil.*
2. *Janssens F. & Peeters A. Soumis. Relationship between nutrient content in the plants and grassland diversity. Plant and Soil.*

En fonction de ces résultats, une méthode simple permettant de juger du potentiel de restauration d'un site a été décrite. Cette méthode, basée uniquement sur un relevé de la végétation en place, permet en outre l'identification des facteurs à changer pour favoriser cette restauration (article 3).

3. *Janssens, F. & Peeters, A. Soumis. Estimation of the potential biological diversity in swards by quick methods. Journal of Vegetation Science.*

Le choix d'un site se fera de préférence sur les sites où le potentiel de restauration est élevé. Il se peut toutefois que la restauration doive se faire absolument sur un site qui ne soit pas adapté (teneurs en phosphore échangeable élevées). Dans ce cas, des méthodes visant à réduire ces teneur dans le sol sont proposées (article 4).

4. *Janssens F., Peeters A. & Bastin O. Soumis. Restoration of soil nutrient contents compatible with the reconstruction of biodiverse swards. Journal of Applied Ecology.*

Enfin, dans de nombreux cas, la restauration d'un couvert herbacé riche en espèces doit se faire par le biais d'un semis. Il est donc impératif de connaître le comportement germinatoire des espèces-cibles. Une étude a donc été réalisée visant à mettre en relation la germination et la survie des plantules de quelques espèces caractéristiques des prairies de fauche avec le statut hydrique ou nutritif (azote, phosphore et potassium) du milieu (article 5).

5. *Janssens F. & Peeters A. Soumis. Germination of 6 species characteristic of species-rich grasslands in relation with nutrient status and water potential. Journal of Ecology.*

VI. PRÉSENTATION DES CINQ ARTICLES COMPOSANT LA THÈSE

A. INFLUENCE DES TENEURS EN ÉLÉMENTS NUTRITIFS DES SOLS SUR LA DIVERSITÉ BOTANIQUE (ARTICLES 1 & 2)

Vers la fin des années septante, les premières théories visant à expliquer la coexistence des espèces au sein de couverts herbacés voient le jour. Al-Mufti et al. (1977) et Grime (1973 & 1979) (figure 1) dégagent clairement une relation entre la production de matière sèche et le nombre d'espèces présentes sur un site. Cette relation a, depuis lors, été confirmée à plusieurs reprises (Wheeler et al., 1982; Wilson et al., 1990).

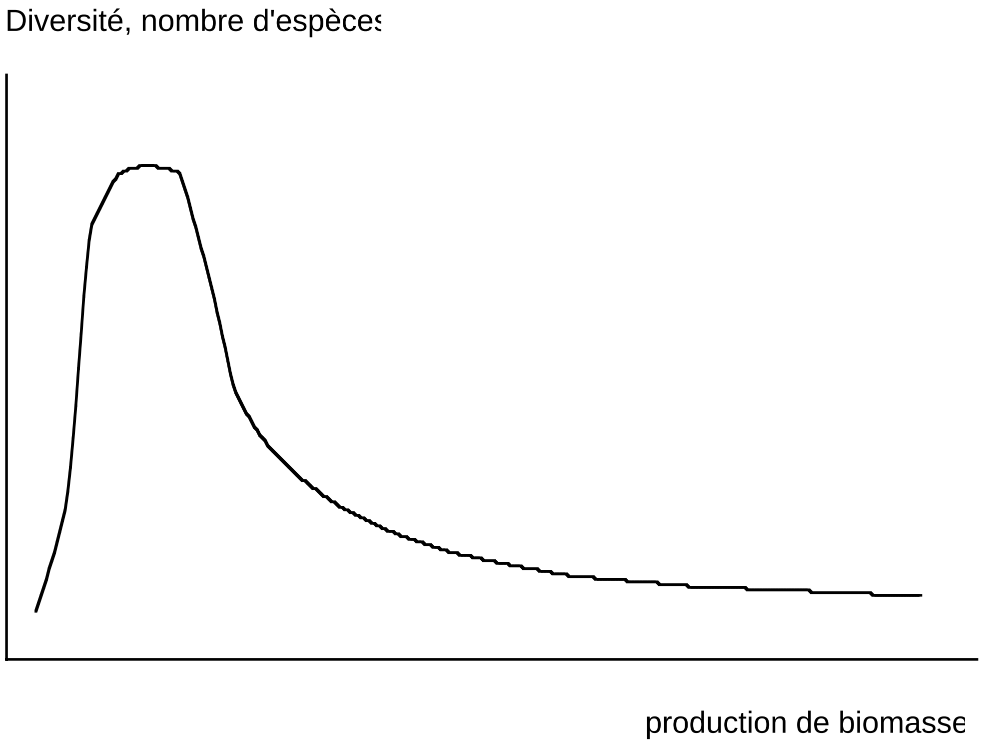

Figure 1. Relation entre la diversité botanique et la production de biomasse (estimée par le nombre d'espèces)

La production de biomasse est définie dans ce cadre comme la quantité de biomasse (végétation vivante et litière) trouvée sur un site au moment du pic de production.

La diversité a été estimée comme étant le nombre d'espèces présentes sur 0,25 m^2.

Cette relation a été établie à partir de relevés effectués sur des sites installés depuis longtemps permettant un certain équilibre entre les conditions pédo-climatiques prévalant à cet endroit et la végétation installée. Les sites ont été choisis afin d'inclure une gamme de végétation herbacée très large (comprenant des prairies pâturées, des prairies de fauche, des praires sèches ou humides) dans le nord de l'Angleterre. Ce modèle est valable pour les régions tempérées. Il est probable que le maximum de diversité trouvé sera différent en altitude par exemple.

Selon Grime (1973), un nombre d'espèces nul est trouvé pour une production de biomasse nulle, c'est-à-dire dans des conditions totalement incompatibles avec la présence d'une végétation herbacée. Dès que les conditions pédo-climatiques s'améliorent et permettent l'apparition de cette végétation, le nombre d'espèces augmente. La couverture herbacée comprend, à ce stade, des espèces adaptées à ces conditions extrêmes (par exemple : une grande pauvreté en éléments nutritifs (landes) ou une teneur élevée en sel (halophytes)). Au fur et à mesure de l'amélioration de ces conditions pédo-climatiques, un nombre de plus en plus important d'espèces peut survivre. Le nombre maximum d'espèces apparaît dans les prairies mésotrophes. Ce nombre maximum d'espèces doit donc son existence à l'action de l'homme qui a défriché les forêts et, petit à petit, enrichi le sol.

Cependant, parallèlement à l'utilisation des fertilisants et à l'intensification des pratiques agricoles en prairies, un autre phénomène est survenu. L'homme, en enrichissant de plus en plus le sol de ces prairies, a favorisé la présence d'espèces à croissance rapide. Ces espèces, comme le ray-grass (*Lolium perenne*) ont peu à peu étouffé les autres espèces. Ceci a eu pour conséquence de baisser le nombre d'espèces présentes de manière drastique (on compte actuellement rarement plus de 5 espèces dans nos pâtures intensives). On pourrait citer, comme autre exemple, l'envahissement de pelouses sèches par des espèces plus eutrophes comme *Brachypodium pinnatum* suite à un enrichissement du sol.

Une autre variable influence la diversité botanique parallèlement aux variables pédo-climatiques : la perturbation de la végétation. Cette perturbation, représentée ici par le nombre de coupes, ne permet qu'une faible diversité botanique lorsqu'elle est nulle. L'application d'un pâturage ou de coupes de foin empêchent le dépôt d'une nécromasse ou l'apparition de ligneux. Ceci permet au milieu de rester ouvert et d'être favorable à l'arrivée et au maintien d'un grand nombre d'espèces végétales. Lorsque ces coupes sont trop nombreuses, la diversité botanique décroît car seules quelques espèces adaptées peuvent survivre.

Suite à l'établissement de ces relations, plusieurs études ont mis en évidence l'effet dépressif d'une fertilisation azotée sur la diversité botanique (Elisseou et al., 1995; Marrs, 1993; Mountford et al., 1993; Tallowin et al., 1994; Willems et al., 1993) (figure 2). Cette fertilisation azotée est effectivement un paramètre essentiel influençant la production de biomasse.

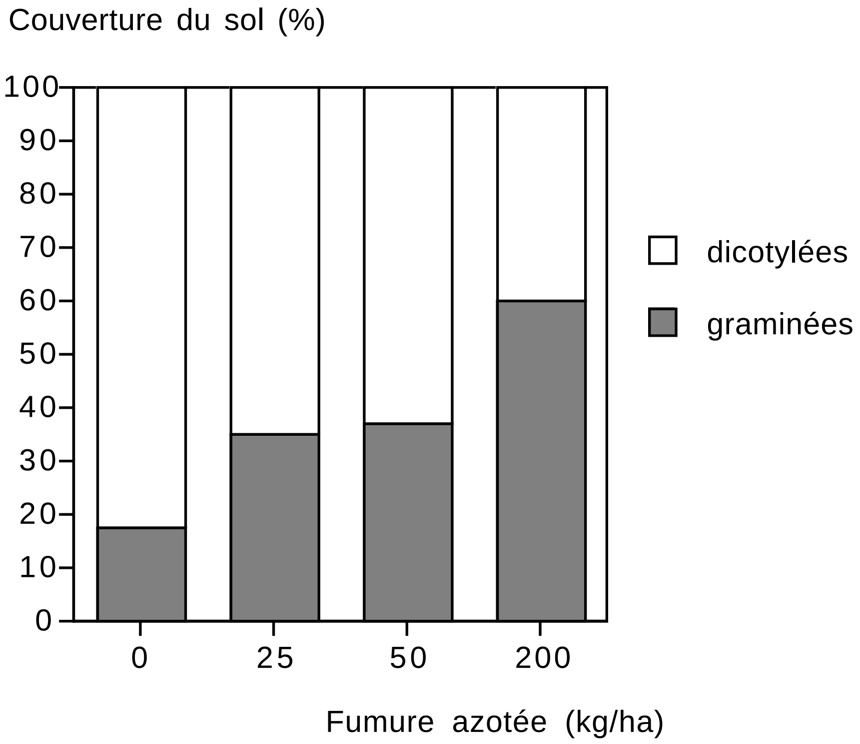

Figure 2. Relation entre la proportion de surface couverte par les graminées en fonction de la fumure azotée (d'après Tallowin et al., 1994)

Cet effet fut alors interprété par la compétition interspécifique; de grandes quantités d'azote permettant aux espèces eutrophes d'exprimer pleinement leurs capacités supérieures de croissance. Les autres espèces, à croissance plus lente, disparaissent alors par manque de lumière (Tilman, 1990).

Ces études se sont particulièrement focalisées sur l'azote, délaissant les autres nutriments. Nous avons donc entrepris, dans le cadre de ce travail, la recherche d'autres éléments du sol en relation avec la diversité botanique.

Le premier article présente le résultat de cette recherche. 281 prairies ont ainsi été analysées tant du point de vue botanique que pédologique. Une relation entre le phosphore échangeable des sols et la diversité botanique a été mise en évidence. La diversité botanique a été exprimée à l'aide de trois paramètres.

Le premier paramètre prend en compte le nombre d'espèces. Contrairement à Grime (1973 et 1979), ce nombre d'espèces, tout comme les autres paramètres, a été établi à partir de relevés effectués sur des surfaces relativement grandes (100 m^2). En effet, Oksanen (1996) a montré le danger d'utiliser des surfaces si petites (0,25 m^2 utilisés par Grime en 1973) dans de telles études de végétation. Selon cet auteur, les facteurs influençant la diversité botanique sur de si petites surfaces relèvent plus de la place occupée par les individus plutôt que des variables pédo-climatiques.

Le second paramètre consiste à calculer un indice représentant un niveau de rareté moyen de la flore présente. Ce calcul se base sur le coefficient de rareté attribué spécifiquement par Lambinon et al. (1992). Il est évident que l'établissement de ces coefficients est relativement subjectif et ne tient pas réellement compte de la vulnérabilité de l'espèce. Même si l'établissement de ces coefficients est régulièrement revu, il n'en demeure pas moins perfectible au niveau de la précision géographique et de la définition de la rareté. En effet, le coefficient attribué pour une espèce est défini pour une large zone. Le fait que l'espèce soit commune dans une partie de cette zone et rare dans une autre partie est ignoré. Le fait que cette espèce soit en régression dans cette zone, et par conséquent vulnérable, n'est pas pris en compte. Quoi qu'il en soit, en l'absence d'un paramètre plus précis, nous avons jugé utile d'utiliser ce coefficient afin d'affiner les résultats obtenus avec le nombre d'espèces. Afin de limiter au maximum les imprécisions, nous avons considéré séparément chaque district phyto-géographique dans nos calculs.

Le troisième paramètre (indice de Shannon) permet de calculer le niveau de dispersion de la communauté végétale. Pour un nombre d'espèces donné, ce paramètre est maximum lorsque toutes les espèces ont un coefficient de recouvrement identique au sein de l'échantillon (Shannon, 1948). Ce paramètre permet donc de mettre en évidence la dominance, par exemple, des espèces à croissance rapide sur les sols riches en éléments nutritifs. Ce paramètre complète donc les informations obtenues par les deux premiers.

Il est apparu que toutes les stations analysées comprenant plus de 5 mg/100 g de phosphore échangeable (extraction à l'acétate d'ammonium et EDTA; Läkanen et al., 1971) comprenait moins de 20 espèces par 100 m^2. Parallèlement, l'indice de rareté et de répartition de la végétation y étaient toujours très faibles. Il aurait été intéressant de quantifier les quantités d'azote disponible dans ces sols. Cette fraction, essentiellement représentée par les nitrates du sol, était trop faible et trop variable pour être mise en évidence avec suffisamment de précision avec les techniques disponibles. Il apparaît a posteriori qu'une technique basée sur une incubation anaérobie permettant d'estimer l'azote potentiellement minéralisable pourrait s'adapter à cette étude. Cette mesure pourra être réalisée plus tard pour compléter les données existantes.

La diversité botanique peut donc être réduite par de grandes quantités d'azote ou de phosphore disponibles. Ces deux éléments peuvent agir séparément, par exemple, en cas de carence ou d'excès d'un des deux éléments. Dans d'autres cas, ces deux éléments pourraient interagir de différentes manières. Dans cette hypothèse, de faibles quantités de phosphore disponible pourraient déprimer l'activité fixatrice d'azote des légumineuses (Hart et al., 1981; Hart, 1982) ou la minéralisation de la matière organique par les microorganismes du sol (Cadish et al., 1994).

Le deuxième article met cependant en évidence le rôle direct du phosphore échangeable des sols sur la diversité botanique. Une relation décroissante entre la teneur en phosphore de la biomasse et le nombre d'espèces trouvées sur 100 m^2 a, en effet, été établie pour des rendements en matière sèche entre 2 et 5 tonnes. Cette relation n'a pu être reproduite lorsque les teneurs en azote de la biomasse étaient considérées.

Il est à noter que, dans cet échantillon, la production de matière sèche n'était nullement liée aux teneurs en phosphore dans la biomasse. De faibles concentrations en phosphore n'ont pas induit une baisse de production de matière sèche. Une explication à ce phénomène réside dans la différence de stratégie des espèces à croissance rapide (eutrophes) ou lente (oligotrophes). Les premières incorporent plus rapidement les nutriments en tissu, cette transformation plus rapide nécessite de plus grandes concentrations en nutriments dans les sols et dans leurs tissus. Les secondes économisent plus les nutriments en diminuant notamment les pertes par sénescence; le rapport entre les concentrations en nutriments dans les tissus et la production de biomasse peut donc être très faible pour ces espèces (Tilman, 1990).

B. MÉTHODES DE DÉTERMINATION DE LA DIVERSITÉ BOTANIQUE POTENTIELLE D'UN SITE (ARTICLE 3)

La restauration des couverts herbacés riches en espèces, pour une efficacité optimale, doit être considérée au sein d'un ensemble plus complexe que l'on nomme communément "le réseau écologique". Ce réseau écologique peut se définir comme l'ensemble des milieux qui permettent d'assurer la conservation à long terme des espèces sauvages sur un territoire. Il implique donc le maintien d'un réseau cohérent d'écosystèmes naturels et semi-naturels, mais aussi d'habitats de substitution, susceptibles de rencontrer les exigences vitales des espèces et de leurs populations (bords de routes) (Melin, 1995).

La considération de cet ensemble implique l'utilisation de grandes surfaces de liaison comme, par exemple, les bords de routes. Les conditions pédo-climatiques régnant à ces endroits ne conviennent pas nécessairement à de tels couverts. Avant d'entreprendre toute action, il est donc utile de juger des possibilités de succès de cette restauration par des méthodes rapides et peu coûteuses.

En fonction de ce qui a été exposé plus haut, des couverts herbacés riches en espèces ne peuvent se maintenir sur des sols trop riches en azote ou en phosphore. Les teneurs de ces éléments dans les sols peuvent bien entendu être analysées par les méthodes classiques. Une analyse de ces teneurs dans la végétation pourrait aussi remplacer ces analyses de sols. Cependant, la variabilité des teneurs en azote en fonction du stade physiologique rend cette analyse difficile à interpréter.

Un inventaire floristique pourrait remplacer avantageusement ces analyses. En effet, elle peut être réalisée rapidement et plus facilement. Ellenberg (Ellenberg et al., 1991) a estimé l'optimum écologique des espèces végétales concernant sept facteurs écologiques, dont l'azote disponible présent dans le sol. Chaque espèce est dotée d'un coefficient indiquant son comportement en fonction de l'azote disponible dans les sols (de 0, pour les espèces n'existant que sur les sols très pauvres en azote, à 9, pour celles trouvées uniquement sur les sols très riches en azote). La somme du recouvrement de chaque espèce multiplié par son coefficient N d'Ellenberg permet de calculer un indice N d'Ellenberg à l'échelle de la parcelle. Cet indice N d'Ellenberg permet ainsi de classer chaque parcelle sur une échelle reflétant un niveau trophique.

Ces indices doivent toutefois être utilisés avec précaution car l'auteur les a établis à partir de sa propre expérience de terrain, sans test expérimental systématique. Ces indices ont, en outre, été établis pour l'Europe Centrale. Thompson et al. (1993) ont cependant montré clairement l'applicabilité de ces coefficients en Angleterre. Cette applicabilité ainsi que l'utilité et la fiabilité de ces indices ont encore été montrées à de nombreuses reprises et dans des conditions fort diverses. Ces études concernent notamment les risques d'eutrophisation dans les prairies humides (Latour et al., 1994), la qualité du sol (Hawkes et al., 1997) et le niveau d'intensivité en prairie (Melman et al., 1988; Hill et al., 1997). Meerts (1997) a d'ailleurs montré que cet indice était un excellent indicateur des teneurs en N et P de 39 espèces graminoïdes et de 84 dicotylées.

Le but de cet article est de proposer une méthode basée sur ces indices d'Ellenberg permettant d'estimer la diversité botanique potentielle d'un site en vue de la restauration de couverts herbacés riches en espèces. Cette méthode implique une bonne connaissance de la flore. Une autre méthode, basée uniquement sur le nombre d'espèces dicotylédones, est enfin proposée. Il s'avère que seuls les sites pauvres en phosphore (moins de 5 mg/100 g) montrent plus de 10 dicotylées différentes par 100 m^2. Cette méthode est certes moins précise que la précédente mais elle devrait s'avérer utile lors d'études préliminaires ou lorsqu'il n'est pas possible de faire des relevés botaniques précis (grandes surfaces, manque de personnel pour reconnaître la flore,...).

C. MÉTHODES À UTILISER POUR DIMINUER LES QUANTITÉS DE PHOSPHORE ÉCHANGEABLE DANS LES SOLS (ARTICLE 4)

Dans le cadre d'une entreprise de restauration de couverts herbacés riches en espèces, il apparaît, suite à ce qui a été présenté ci-dessus, que des sites présentant de hautes teneurs en azote ou en phosphore, devraient être évités. Il est toutefois possible que, dans certains cas, on ne puisse pas choisir. Il sera alors nécessaire d'appliquer des mesures visant à réduire la disponibilité de ces éléments.

Ces mesures devraient essentiellement se focaliser sur le phosphore plutôt que sur l'azote. En effet, ce dernier élément est apporté soit par la fertilisation (il suffit de l'arrêter et d'éviter le ruissellement en provenance de parcelles adjacentes) ou par les retombées atmosphériques. De plus, cet azote disponible, essentiellement sous forme de nitrate, est très mobile dans les sols, il est donc très rapidement lixivié. Des quantités d'azote non négligeables peuvent toutefois être apportées par la minéralisation de la matière organique ou par la fixation symbiotique. Ces quantités apportées par la fixation peuvent être réduites par des quantités de phosphore disponible faibles diminuant l'efficacité de la symbiose. Les quantités d'azote minéralisées dépendent de la quantité de matière organique et du coefficient de minéralisation. Des quantités de phosphore disponible faibles peuvent aussi diminuer ce coefficient de minéralisation. Il est de plus probable que cet élément déprime directement la diversité botanique dans ces couverts.

Il semble donc, à ce stade, très important de diminuer les quantités assimilables de phosphore échangeable avant toute entreprise de restauration de couverts herbacés riches en espèces.

Une première méthode consiste à retirer une couche de sol (étrépage). Les couches supérieures de sols sont en effet plus riches en éléments nutritifs et spécialement en azote organique et en phosphore. Ceci s'explique par la faible mobilité de ces éléments qui entraîne leur accumulation dans les couches superficielles de sol. Cette méthode est bien entendu radicale mais peut difficilement être appliquée partout. Elle s'avére utile dans les réserves naturelles, par exemple, ou dans tout espace vert non agricole.

L'addition d'un substrat plus pauvre (du sable par exemple) pourrait faire l'objet d'une autre méthode, plus facilement applicable. Elle pourrait avantageusement être utilisée sur les bords de routes avec les terres extraites lors de la construction.

Ces méthodes nécessitent la destruction de la flore présente. Il peut, dans certaines conditions, s'avérer avantageux de conserver la flore en place et de l'améliorer par une gestion adaptée avec, éventuellement, un apport artificiel d'espèces. Dans ce cas, deux autres méthodes peuvent être présentées.

La première, largement utilisée, se base sur l'extraction des nutriments par les végétaux. La coupe de la biomasse et l'exportation de celle-ci appauvrit le sol. Cependant, il a été montré (Marrs, 1993) que cette méthode nécessite souvent une application durant plusieurs décennies avant de s'avérer vraiment efficace. Ceci concerne de nouveau essentiellement le statut phosphorique du sol. Les quantités exportées de cet élément sont très faibles (10-20 kg/ha) comparativement aux quantités totales, susceptibles d'être mobilisées à plus ou moins long terme par les plantes (plusieurs tonnes par ha).

La seconde utilise un phénomène naturel impliquant des oxydes de fer. Ces oxydes de fer, présents naturellement dans les sols, adsorbent les anions phosphates plus ou moins fortement et les soustraient à la solution du sol, les rendant ainsi moins accessibles à l'absorption végétale. Le principe de cette méthode consiste simplement à accentuer un processus tout à fait naturel. Une étude est par ailleurs ciblée sur l'augmentation de la réactivité de ces oxydes vis-à-vis des ions phosphates afin de n'en utiliser qu'une petite quantité (2-3 %) par rapport aux quantités déjà présentes dans les sols (plusieurs dizaines de tonnes par hectare). Les oxydes de fer utilisés sont issus de processus sidérurgiques permettant ainsi de combiner le recyclage de déchets industriels et la protection de l'environnement. Ils ne sont, bien entendu, pas voués à être épandus massivement sur les terres agricoles. Ils pourraient par contre s'avérer très utiles sur des anciennes parcelles agricoles ainsi que sur des sites jouxtant des sites agricoles (bords de routes, parcs publics, jardins privés).

Ces oxydes, qui se sont révélés purs mais cristallins (hématite), ont été testés en solution et sur le terrain. Les quantités de phosphate en solution au laboratoire ou de phosphore du sol extractible à l'eau ont été significativement diminuées par l'adjonction de ces oxydes. Les méthodes d'extraction chimique du phosphore dans les sols n'ont par contre pas montré de résultats marquants. Des traitements chimiques ont été appliqués sur ces oxydes afin d'en améliorer l'activité. Puisque dans notre cas, la surface spécifique semblait être l'élément limitant (Borggaard et al., 1990), son accroissement a été le principal objectif de ces traitements.

Le but de cet article est de faire le point sur les différentes méthodes disponibles pour restaurer des sols en vue de les rendre aptes à supporter des couverts herbacés riches en espèces. Puisque les travaux précédents ont permis la mise en évidence de l'importance du phosphore échangeable dans les sols, une méthode adaptée spécifiquement à ce problème y est présentée.

D. GERMINATION DES GRAINES ET SURVIE DES PLANTULES EN SOLUTION NUTRITIVE DE QUELQUES ESPÈCES "MODÈLES" DES PRAIRIES DE FAUCHE EN FONCTION DU POTENTIEL HYDRIQUE ET DES TENEURS EN AZOTE, PHOSPHORE ET POTASSIUM (ARTICLE 5)

Des conditions pédologiques défavorables ne constituent pas le seul élément susceptible d'empêcher la restauration de couverts herbacés riches en espèces. Les espèces constituant le couvert souhaité peuvent elles-mêmes manquer. Naturellement, ces espèces peuvent être présentes sur le site à restaurer dans la banque de graines ou y arriver par la pluie de graines. C'est le cas, par exemple, des espèces colonisant rapidement les clairières forestières (*Epilobium angustifolium*). Les graines de beaucoup d'espèces messicoles comme le coquelicot (*Papaver rhoeas*) peuvent persister, dormantes, dans les sols pendant plusieurs décennies avant de germer et de recoloniser un champ cultivé.

Plusieurs études ont toutefois montré que la banque de graines ou la pluie de graines des espèces caractéristiques des prairies riches en espèces étaient insuffisantes dans un tel but (Dutoit et al., 1995; Mc Donald et al., 1996). En effet, les semences de nombreuses espèces intéressantes ne peuvent survivre plus de 5 ans dans le sol (Bakker et al., 1991). De plus, des études menées en Angleterre (Kirkham et al., 1997; Tallowin, non publié), en Espagne (Fillat, non publié) et en Hollande (Bakker, non publié) ont montré que le nombre d'espèces intéressantes présentes dans la banque de graines était inversement corrélé au niveau de richesse du sol en éléments nutritifs (notamment au contenu en azote et en phosphore disponibles du sol). Le drainage, souvent réalisé dans notre pays, s'est avéré dans cette dernière étude particulièrement préjudiciable. Ceci rejoint les travaux de Cavers et al. (1989) qui ont montré que la présence d'oxygène en concentration importante avait un impact particulièrement négatif sur la survie des semences dans les sols. Dans la plupart des cas, suite à un drainage ou un apport de fertilisant azoté ou phosphaté, les semences d'espèces intéressantes disparaissent au profit de celles d'espèces pionnières. Le retour aux conditions initiales (suppression de la fertilisation ou du drainage) n'assure absolument pas la régénération de la banque de graines à court terme. Bakker (non publié) a obtenu un changement significatif de la banque de graines seulement 20 ans après la cessation de fertilisation et l'application d'un programme de gestion adapté. Ceci rejoint les résultats obtenus outre-Atlantique par Major et al. (1966). Marrs (1993) a rassemblé ces observations dans une théorie ("catastrophe model") où il explique la décroissance de la richesse en espèces d'un site en fonction d'une fertilité croissante du sol (figure 3). Cette décroissance se fait progressivement entre le point A et B, au-delà de ce point, une chute brutale est observée jusqu'au point C. La décroissance progressive reprend du point C au point D, laissant la place à une chute brutale, etc. La restauration de la richesse en espèces par réduction de la fertilité du sol pourra se réaliser entre le point D et le point C. Cependant le passage de C à B ne pourra se réaliser que par une autre méthode plus radicale (apport d'espèces).

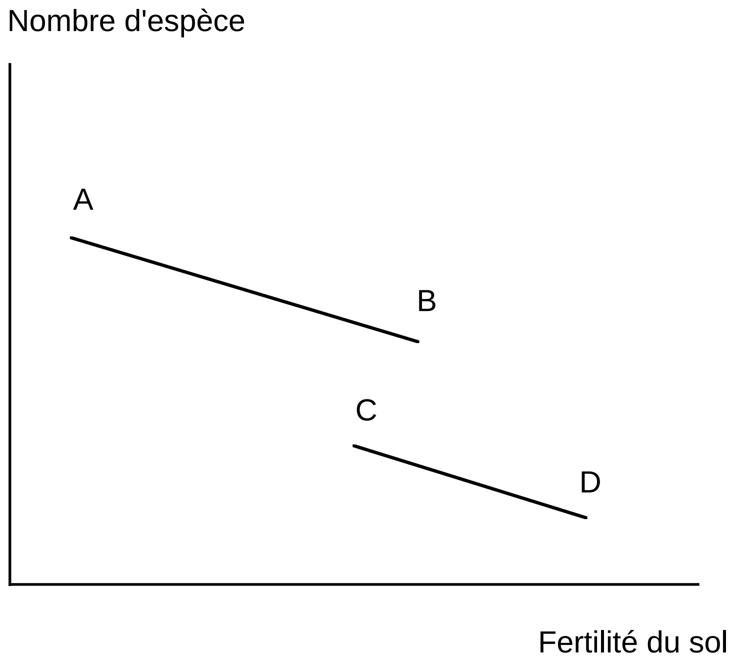

Figure 3. Relation entre le nombre d'espèces et la fertilité du sol (modèle catastrophe (Marrs, 1993))

La restauration rapide d'un site ne semble pas pouvoir être uniquement basée sur l'apport de semences par la pluie de graines, même si le site se situe dans une région où les couverts herbacés riches en espèces abondent (Bakker et al., 1996). Ceci s'explique probablement par un apport trop faible et un établissement difficile (faible germination, mortalité élevée des plantules). De plus, Kirkham (1996) a montré l'effet néfaste de teneurs en azote élevées dans les sols sur la production de graines par les espèces typiques de ces milieux.

A la lumière de ces études, il apparaît que la restauration des couverts herbacés riches en espèces ne peut être réalisée, dans beaucoup de cas, que par un semis ou un repiquage des espèces manquantes (Dutoit et al., 1995; Hutchings et al., 1996 a & b). Afin de préserver l'originalité génétique des populations, les semences ou les plants utilisés doivent provenir de populations issues de la même région (Lambinon, 1994 & 1997).

Peu d'études ont été réalisées à ce jour sur la germination et la survie des plantules d'espèces de ce type. Il semble que les plantules survivent difficilement dans les couverts herbacés et ce, surtout suite à un semis réalisé au printemps (Janssens et al., 1996). Ceci s'explique essentiellement par les conditions de sécheresse survenant durant l'été. La compétition intense entre les plantes adultes due à cette sécheresse est très préjudiciable pour les plantules (Tallowin, comm. or.). Les meilleures conditions de survie semblent se situer dans les végétations régulièrement coupées suite à un semis en fin de saison (août-septembre). Ces conditions permettent une atténuation des variations de température et d'humidité au niveau du sol (Hutching et al., 1996 b). Ce sujet, extrêmement large et complexe, nécessite encore beaucoup d'études. De plus, la survie des plantules est subordonnée à la germination des espèces semées. C'est pour cette raison que nous avons essentiellement concentré le sujet de cet article sur la germination des semences d'espèces caractéristiques des prairies de fauche riches en espèces. Cette étude est certainement aussi complexe que la précédente. Nous avons donc focalisé l'étude sur les facteurs mis en évidence dans le début de ce travail comme influençant la végétation herbacée dans les couverts riches en espèces. Nous avons donc étudié l'influence des éléments nutritifs majeurs (azote, phosphore et potassium) sur la germination d'espèces caractéristiques de ces milieux. Parmi celles-ci, six espèces ont été choisies en fonction de tests préalables établissant leurs relatives facilités de germination. L'influence de ces éléments nutritifs peut être fortement altérée par un changement de potentiel hydrique (Goudey et al., 1988; Copeland et al., 1995). Il était donc particulièrement important d'inclure ce dernier facteur dans notre étude. La dormance des semences n'a pas été approchée dans ce travail. Il aurait, en effet, fallu aborder des thèmes extrêmement complexes comme les conditions de maturation sur la plante-mère, les dormances secondaires spécifiques,... Les semences utilisées sont donc celles qui peuvent être obtenues dans le commerce. Les conditions de maturation et de stockage (température de 2-3°C, obscurité) ont été identiques pour les 6 espèces testées. Aucune action visant à modifier les états de dormance spécifique des graines fournies n'a été réalisée.

Très peu d'études de ce genre ont été réalisées à ce jour, il existe donc peu de points de comparaison possibles. Grime (1981) a cependant réalisé une étude très complète sur la germination de 403 espèces dans différentes conditions. Cette étude peut servir de base comparative pour le comportement germinatoire de ces espèces, mais les conditions dans lesquelles notre expérience a été réalisée (sécheresse, teneurs en nutriments) n'ont pas été testées par Grime. Selon Egley et al. (1985), la plupart des ions inorganiques, exception faite des ions nitrates, n'ont pas d'influence sur la germination. Les mécanismes d'action de ces derniers sur la germination demeurent inconnus. A faible dose, il a été montré depuis longtemps qu'ils pouvaient promouvoir la germination (Lehman, 1909). Plus récemment, il est apparu qu'ils pouvaient se substituer à la lumière ou entrer en synergie avec elle pour lever la dormance (Karssen et al., 1993; Tallowin, non publié). A forte dose, ces ions ont parfois agi négativement sur la germination. L'origine de cette inhibition n'est pas connue, elle peut être due à une augmentation de pression osmotique et donc agir de la même façon que la sécheresse; ces ions pourraient aussi pénétrer dans la graine et y induire des perturbations physiologiques (Karssen et al., 1993).

Le but de ce dernier article est donc la mise en évidence des effets des nutriments (azote, phosphore et potassium) en différentes quantités sur la germination d'espèces typiques des prairies de fauche de nos régions. Une étude de l'effet osmotique de ces facteurs a aussi été réalisée.

VII. RELATIONSHIP BETWEEN SOIL CHEMICAL FACTORS AND PLANT DIVERSITY IN GRASSLAND

A. SUMMARY

Many studies carried out during these last few years have focused on the factors influencing plant diversity in species-rich grasslands. This is due to the fact that these ecosystems, among the most diversified in temperate climates, are extremely threatened; in some areas, they have almost disappeared. The re-establishment of these habitats implies to know the living conditions of the associations to be recreated. Very often, the typical species of these communities have become so rarefied that the seed bank or the seed rain are not sufficient to recreate the plant community. Most of the time, to achieve the restoration of these communities, they have to be totally recreated by sowing. For the restoration or the maintenance of the community, the soil chemical characteristics have also to be appropriate or if not modified. This research tends to establish a relation between some soil chemical factors and the plant diversity of a great number of stations.

This research has illuminated the relationship between soil extractable phosphorus and potassium and plant diversity. Over 5 mg of phosphorus per 100 g of dry soil (acetate + EDTA extraction), no station containing more than 20 species per 100 m^2 has been found. The highest number of species is found below the optimum content of the soil for plant nutrition (5-8 mg P/100g).

Concerning the potassium, the highest number of species is found at about 20 mg/100g, a value corresponding to an optimum content of the soil for plant nutrition. High potassium contents, in opposition to phosphorus contents, are thus compatible with high values of diversity. Other factors (i.e. pH, organic matter, total nitrogen and calcium) do not show so clearly a relation with plant diversity. Excess of N-NO3 is known for its negative effect on the diversity of plant communities. In these environments, apart from the atmospheric deposits which can be important in some areas, N-NO3 is derived mainly from the symbiotic fixation of atmospheric nitrogen by legumes as well as from the mineralization of the organic matter of the soil. It is possible that, when in small quantities, the available soil

phosphorus could be a limiting factor of the N-NO3 supply by these two sources. In this hypothesis, nitrogen would remain the main element limitating plant diversity but its availability would be controlled by phosphorus.

B. INTRODUCTION

The grassland management intensification of these last decades has considerably reduced the importance of diversified swards in West-Central Europe (Baldock, 1990). The main factors affecting grassland diversity are the cut regimes (especially silage involving early cuts) and the increase of fertilizer use (especially nitrogen and phosphorus). The grassland flora, which is the most species-rich ecosystem in the concerned area, has become more and more simplified to eventually be reduced, in some cases, to its lowest value : the monospecific sward. The maintenance of some plant communities in nature reserves has nowadays become a common practice to slow down this evolution. This solution is certainly very useful but it is insufficient. The threatened plant communities, kept in reserves, are indeed totally isolated one from each other and this consequently annihilates the gene exchange necessary to ensure their subsistance (Spellerberg, 1991).

Some areas could however create a link between these communities (ecological network) : roadsides, public parks, industrial zones and especially a part of agricultural lands could be converted into areas which would be more adapted to wild flora and fauna. The agri-environmental program of the Common Agriculture Policy (directive 2078/92) has recently been implemented with a view to taking this opportunity. It concerns the agricultural lands, marginal or not, which could be used for an ecological management (herbaceous field margins, hedges, late cuts, ...) on the basis of the payment of a premium to the farmer.

The aim of reconstruction or restoration of species-rich swards in these lands, implies that the seeds of the species of the target community still exist in the seed bank or are brought by the seed rain. These seeds must be viable and the seedlings produced by these seeds must be able to survive and develop in the sward. However, it seems that the seed supply is insufficient in most cases. Indeed, the rarefaction of diversified grasslands makes the seed rain insignificant. Moreover, most of the grassland species seeds have a short persistance in the soil (Mc Donald et al., 1996; Thompson et al., 1997). This means that, in many cases, the restoration of an association will have to be carried out by sowing. So, we need to know not only the conditions in which the community germination can occur (light, temperature, humidity,...) but also the conditions (particularly soil chemical characteristics and cutting regimes) which have to be gathered in order to ensure the establishment, the survival and the persistence of the requested plant community.

Al-Mufti et al. (1977) and Grime (1979) indicate that there appeared to be a humped-back relationship between biomass and number of species in herbaceous communities. Since then, this relation has been demonstrated as well by various authors (Wheeler and Giller, 1982; Wheeler and Shaw, 1991; Wilson and Shay, 1990). On the other hand, some authors like Vermeer and Verhoeven (1987) did not find the typical humped curve; it is possible their plots were too small (0.4 m^2). Oksanen (1996) has indeed assumed that the humped-back relation can be due to small plot size independantly to any ecological reason. We obtained this humped-back relation with measures of dry matter yields and species numbers on larger plots (100 m^2), this will appear in a subsequent paper.

Nevertheless, it can be argued that the maximum biomass depends on the production of the community and thus on the fertility of the soil. Therefore, a humped-back relationship between number of species and soil fertility can be expected. This paper aims to find out this relationships.

Several studies have shown that when nitrogen fertilizers were spread (even at small quantities), the species number decreased (Elisseou et al., 1995; Marrs, 1993; Mountford et al., 1993; Tallowin et al., 1994; Willems et al., 1993). It could be thought that the cancellation of nitrogen spreading is sufficient to reach a level in available nitrogen in the soils low enough to allow species-rich communities. However, the cessation of the N fertilization is not sufficient in some cases. The N-NO3 and N-NH4 soil contents have indeed other origins (mineralization of organic matter, symbiotic fixation of atmospheric nitrogen by legumes) which could be important.

Other soil chemical characteristics such as phosphorus or potassium could however decrease species richness. These soil nutrients are indeed present at high levels in some agricultural soils because they are widely used. Other characteristics are related to soil pH or organic matter. These elements are moreover closely related to the N, P, K availability in the soils.

This study aims to demonstrate the relation between the soil chemical characteristics and plant diversity in West-Central European grasslands. The analysis concerns a large number of fields spread over Europe and will allow more precise definition of Grime's model. Moreover, this will provide a more precise targeting criterion in relation to soil nutrient availability for reconstructing floristically diverse grasslands.

C. MATERIAL AND METHODS

The study has been carried out in the temperate region of Western and Central Europe (Belgium, the Netherlands, Great-Britain, Spain and Luxemburg) (figure 1). The sample includes 281 stations with a wide range of soils regarding their nutrient content, their pH, their texture and their water supply (table 1).

Series	Country	Location	Soil type	n
1	Belgium	"Viroin-Hermeton" Park overlapping Ardenne and Famenne region	Ardenne : heavy loamy soils with a good natural drainage, or clayey soils with moderate to poor natural drainage or loamy to loamy-stony soils acid , very wet. Famenne : mainly calcareous and clayey and are well drained.	129
2	Belgium	"les Bulles" in Gaume region	clayey with imperfect drainage causing great hydrological variations.	31
3	Belgium	Herve region	mainly loamy, wet, weakly acid and rich in nutrients (very intensive agriculture)	26
4	Belgium	Walloon Brabant	loamy and rich in nutrients (intensive agriculture)	13
5	Belgium	Bastogne area, located in the Middle-Ardenne.	loamy-clayey, slightly acid and relatively rich in nutrients	7
6	Belgium	Bastogne area, located in the Middle-Ardenne	like series 5 but chosen out of agricultural area, in wet (*Filipendula* or *Comarum* Community) or dry (Violo-Nardion) grasslands	8
7	The Netherlands	Wageningen	Sandy or clayey, wet to dry	4

8	The Netherlands	Groningen	acid and wet peat soils - 1 to 4 - or dry and humic - 5 to 8	8
9	Great-Britain	Devon-North Wyke	impervious clay soils	8
10	Spain	Pyrenees-Jaca	morainic deposits, slightly acid	4
11	Luxembourg	Gütland	wet, clayey soils, with a gley horizon, neutral to slightly acid	43

Table 1. Description of the sites. (n = number of sites - total = 281)

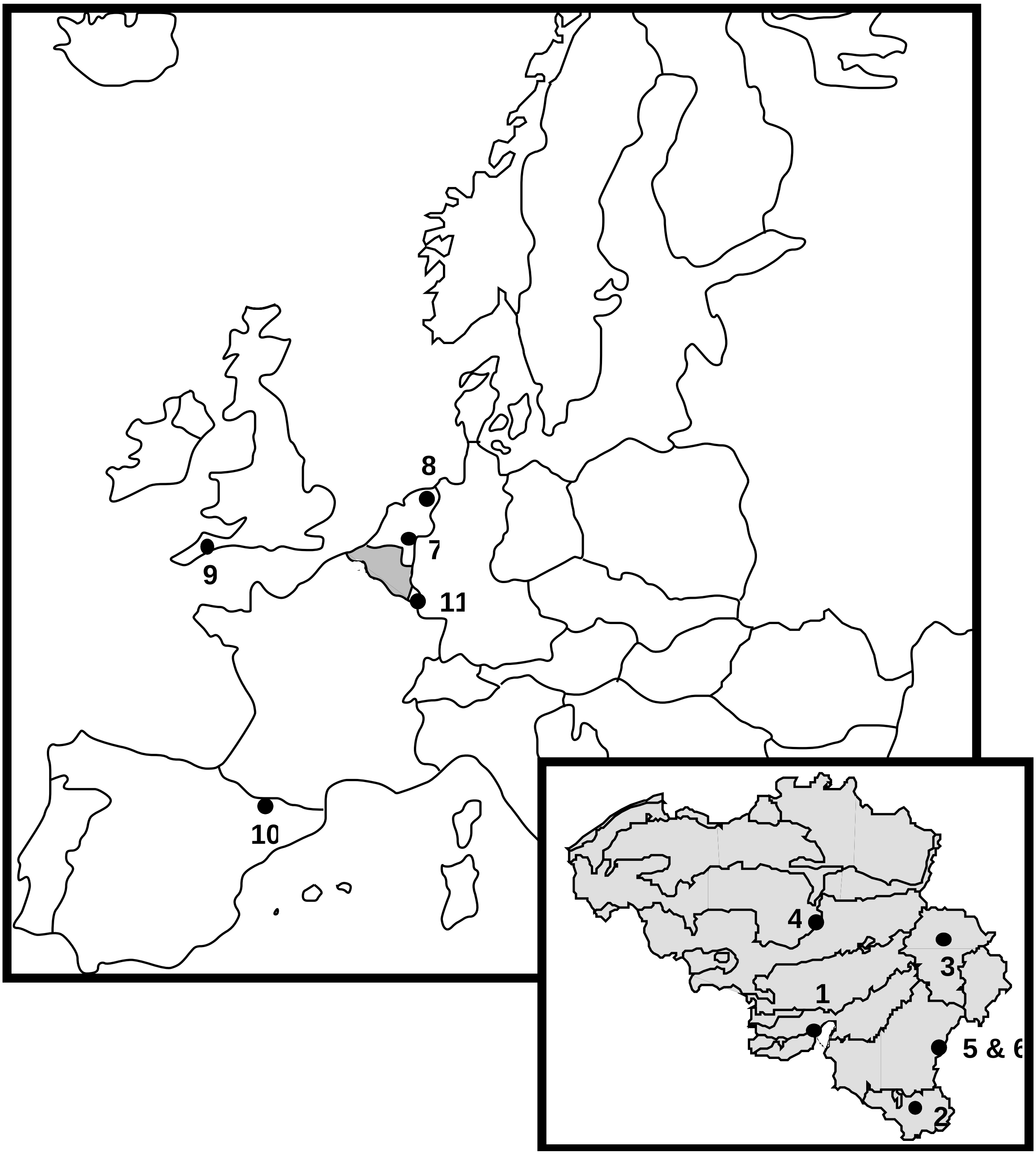

Figure 1. Localisation of the sites.

All the stations are old permanent grasslands. Their management, very different from each other have been applied for many years.

The soils of each of these stations have been sampled (20 samples of 2 cm diameter) between 0 and 15 cm depth and analysed for : pH(H_2O), organic matter (O.M.) (oxydable carbon : Walkley and Black, 1934), total nitrogen (Nt) (Kjeldahl), exchangeable nutrients : Phosphorus and potassium (extraction by acetate-EDTA : Läkanen and Erviö (1971); Cottenie et al. (1975); measure with atomic absorption spectrophotometer except for phosphorus (colorimeter)) (table 2). The units used to present the soil nutrient are mg/100 g dry soil, except for nitrogen : % dry soil.

	Series 1	Series 2	Series 3	Series 4	Series 5	Series 6	Series 7	Series 8	Series 9	Series 10	Series 11
pH	6.56	6.33	6.33	-	6.60	5.35	5.32	5.55	5.14	7.18	6.58
	0.61	*0.24*	*0.17*	-	*0.10*	*0.20*	*0.12*	*0.09*	*0.64*	*0.34*	*0.56*
OM	6.87	10.05	6.08	4.45	6.60	8.85	22.64	-	36.08	7.39	6.81
	2.97	*1.46*	*0.85*	*1.55*	*0.48*	*3.37*	*11.36*	-	*20.49*	*3.03*	*1.68*
Nt	-	0.62	0.45	0.30	0.38	0.60	0.84	0.70	0.87	0.53	0.45
	-	*0.11*	*0.08*	*0.12*	*0.03*	*0.12*	*0.40*	*0.37*	*0.60*	*0.19*	*0.16*
P	4.18	3.03	14.32	13.28	13.79	2.80	2.73	2.38	0.79	1.18	0.63
	5.87	*0.55*	*3.75*	*7.65*	*3.44*	*1.18*	*3.52*	*0.91*	*0.67*	*0.21*	*0.57*
K	18.99	14.05	27.37	39.24	28.43	10.58	9.22	11.86	15.67	26.05	11.59
	10.44	*2.86*	*12.78*	*26.62*	*9.98*	*2.97*	*4.30*	*5.66*	*5.58*	*8.64*	*4.51*
n	129	31	26	13	7	8	4	8	8	4	43

Table 2. Analyses achieved (mean and standard deviation (italic) and numbers of stations (n) studied in each series).

Vegetation has been analysed by the Braun-Blanquet method (Braun-Blanquet, 1932) on 100 m^2 areas (10 m x 10 m).

A rarity index has been calculated by using a relative rarity coefficient of each plant per region. These rarity coefficients have been established by attributing a figure value to the appreciations of Lambinon et al. (1992) (from 0 = very common to 5 = very rare). The rarity index has been calculated for each plot, according to the cover, expressed in percentages, of each species (rarity weighted index). This index was only calculated for Belgium and Luxembourg (91% of the plots).

Rarity index : $\sum_{i=1}^{s}\left(\frac{P_i * 100}{\sum_{i=1}^{s} P_i} * Cr_i\right)$

The Shannon's diversity (Shannon, 1948) has also been calculated : $-\sum_{i=1}^{s}(P_i * \log_2 P_i)$.

Where **P_i** is the proportion of the i^{st} species

Cr_i relative rarity coefficient of the i^{st} species

s is the number of species

D. RESULTS

The number of species shows a humped-back curve with soil extractable phosphorus (figure 2). There is no station with more than 20 species per 100 m^2 on soils containing more than 5mg/100 g of extractable phosphorus. The maximum of species is achieved at 4 mg/100 g of extractable P.

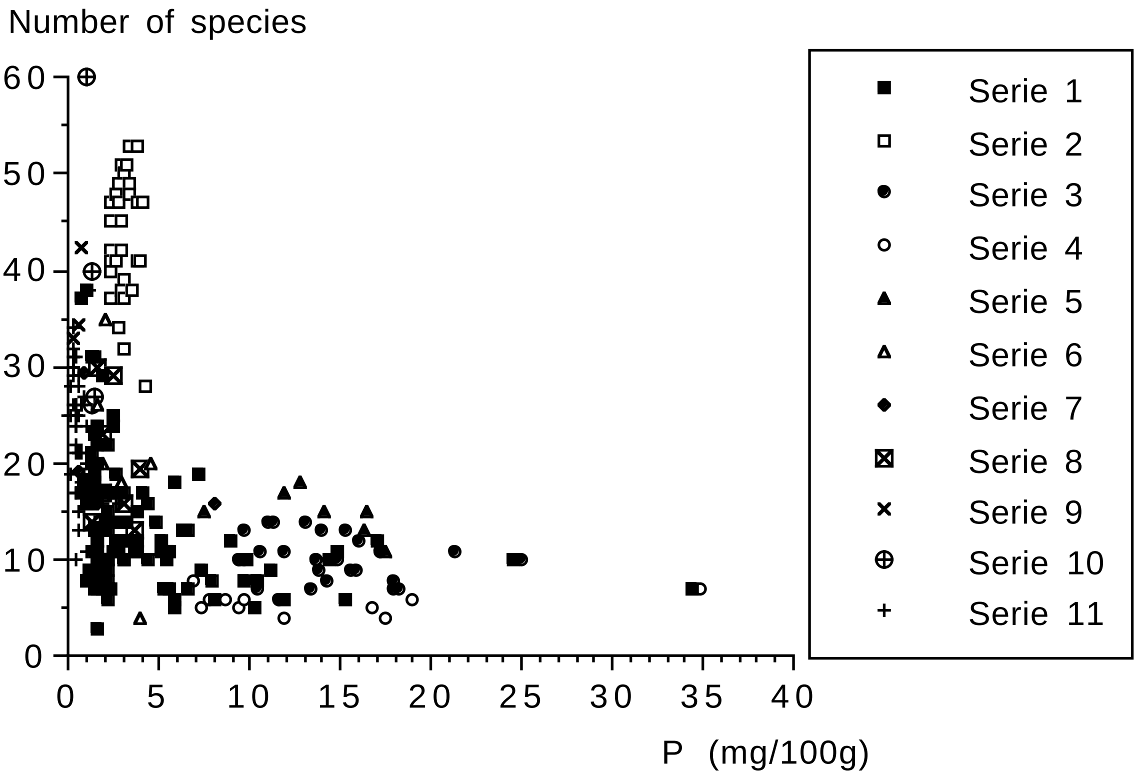

Figure 2. Relation between the soil extractable phosphorus and the number of species per 100m2.

The same relation can be observed with the soil extractable potassium (figure 3). The maximum number of species appears between 15 and 20 mg/100 g extractable potassium.

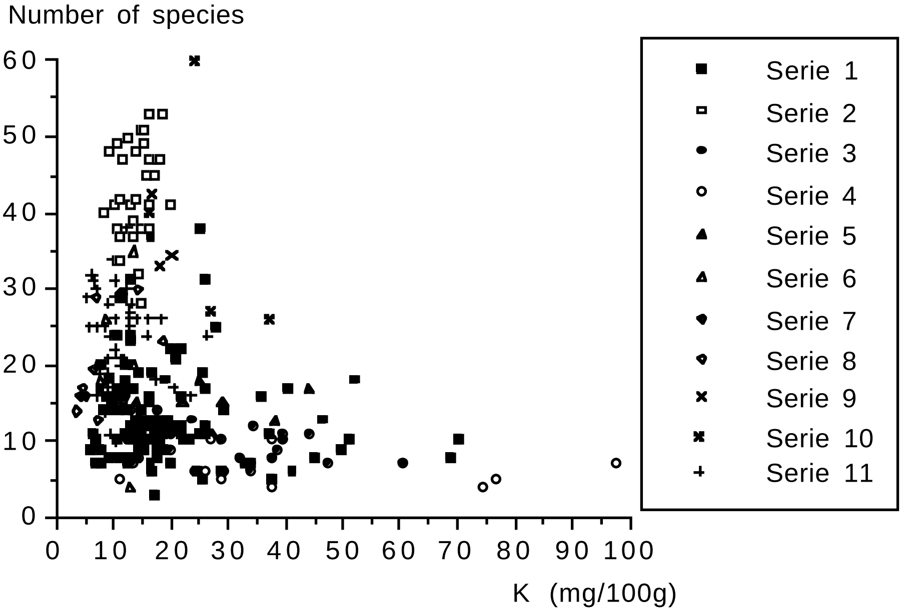

Figure 3. Relation between the percentage of the soil extractable potassium and the number of species per 100 m^2.

The other analysed elements do not show the same clear limit above which no species-rich grassland was observed (figures 4,5 & 6).

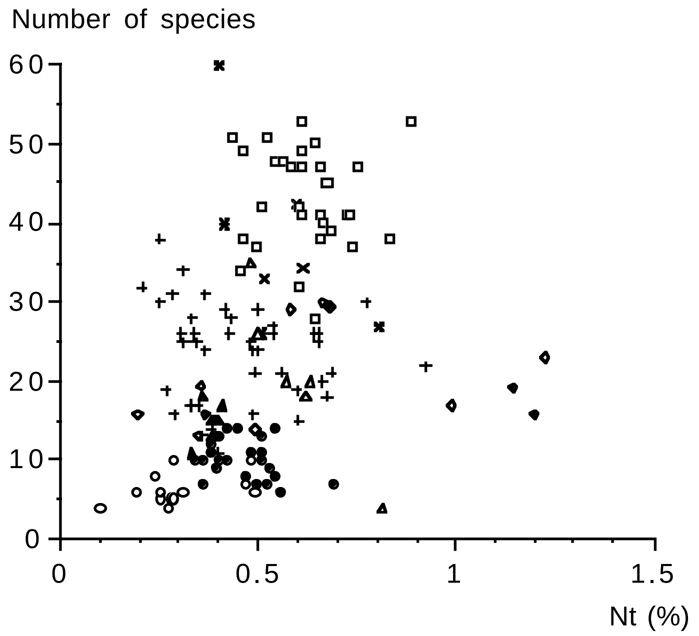

Figure 4. Relation between total nitrogen and the number of species per 100 m^2*. (Legend : see figure 3)*

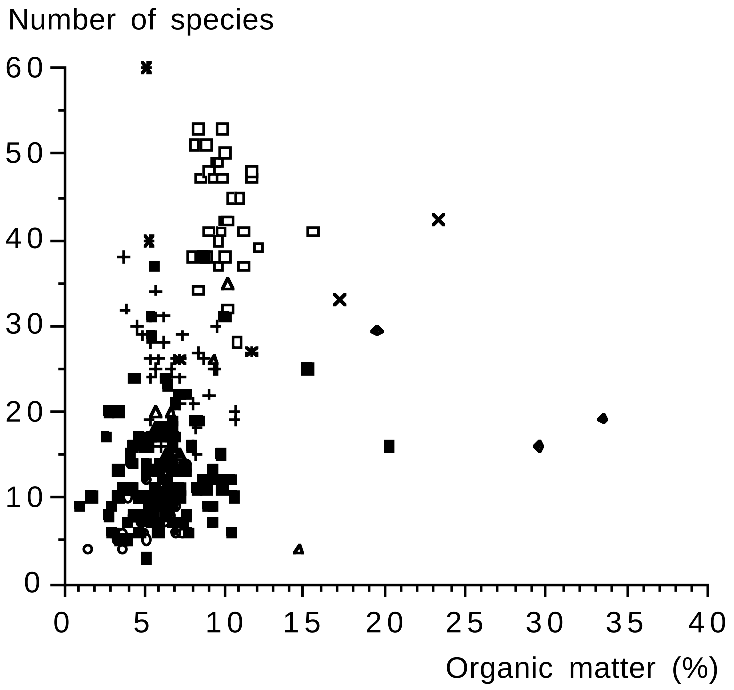

Figure 5. Relation between organic matter and the number of species per 100 m^2*. (Legend : see figure 3)*

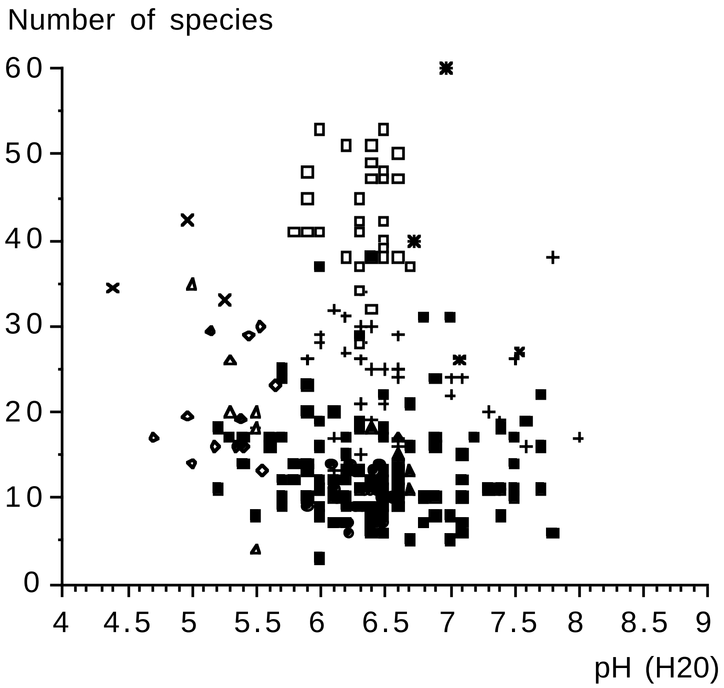

Figure 6. Relation between pH (H_2O) and the number of species per 100 m^2. (Legend : see figure 3)

The rarity index also decreases with phosphorus (figure 7). It can be noted that over 5 mg/100 mg of extractable phosphorus, rarity indices higher than 0.7 have never been observed. The same relation can be reproduced with the Shannon's index (figure 8); this index highly decreases as well for quantities exceeding 5 mg/100 mg of extractable phosphorus.

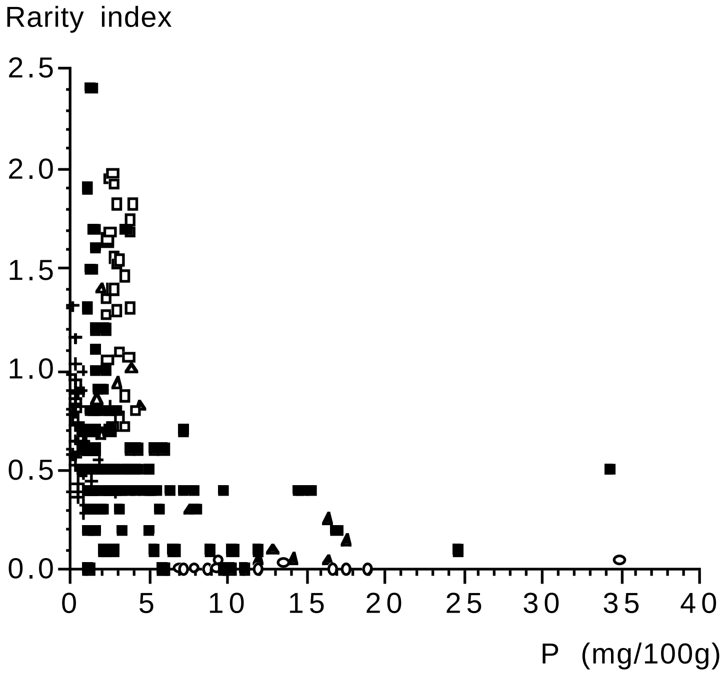

Figure 7. Relation between the soil extractable phosphorus and the rarity index. (Legend : see figure 3)

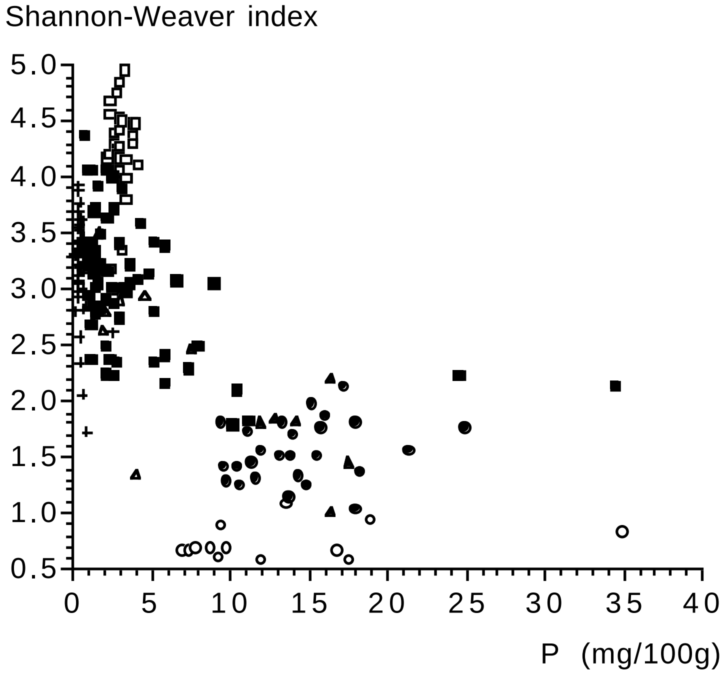

Figure 8. Relation between the soil extractable phosphorus and the Shannon index.

(Legend : see figure 3)

This level also constitutes the minimum level for observing more than three species classified as rather rare to very rare (figure 9) or more than 5% covered by this class of species (figure 10).

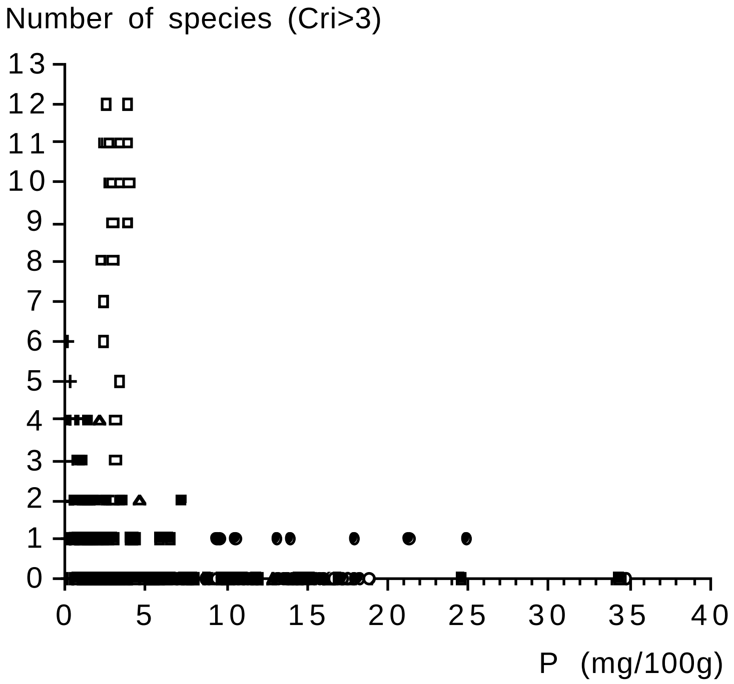

Figure 9. Relation between the soil extractable phosphorus and the number of rather rare to very rare species per 100 m^2*. (Legend : see figure 3)*

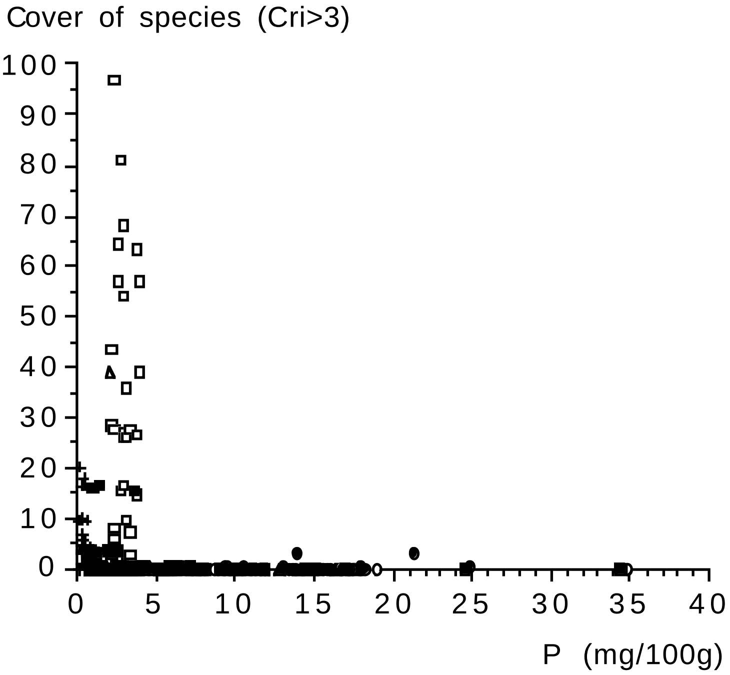

Figure 10. Relation between the soil extractable phosphorus and the cover of rather rare to very rare species. (Legend : see figure 3)

E. DISCUSSION

The humped-back curve between standing biomass and species number found by Al-Mufti et al. (1977) and Grime (1979) can be obtained by replacing standing biomass with major soil nutrients like nitrogen, phosphorus and potassium (figures 2, 3 and 4). The envelope curve shape, representing the maximum number of species when only the X-axis variable (N, P or K) is limiting, is indeed consistent with those found by these authors. The soil fertility represented here by the major nutrients has a similar shape than humped-back curve. The maximum of species number reached at a very low level of available phosphorus, is so close to the Y axis as to be indistinct. The optimum for total nitrogen is about 0.5% dry soil. The relation with the other soil characteristics measured (organic matter, pH) is not so clear indicating a more complex interaction with the species number. These factors are indeed major factors influencing the availability of N, P and K.

Within the framework of the reconstruction or the conservation of grassland floristic diversity, high contents of soil extractable phosphorus seem to be, in many cases, an obstacle of great importance. Indeed, over 5 mg/100 g dry soil, there was no observation of grasslands containing more than 20 species per 100m^2 (figure 2) (and especially the grasslands, which can be considered as diversified, containing more than 30 species) and their maximum rarity index does not exceed 0.7 (figure 7) (intermediary coefficient between the very common to common species class and the common species class). Over this limit, the species registered as rather rare to very rare represent less than 5% of the total cover and do not exceed the total number of 3. Therefore, there is a maximum net limit for the soil content of extractable phosphorus and that over this limit, the probability to observe a diversified sward is very low. Most of the agricultural lands have very often phosphorus contents higher than 5 mg/100 g (table 2). If a species-rich community has to be reconstructed, it seems necessary, at least on an organic matter-rich soil, to choose soils with extractable phosphorus contents below this limit or to decrease these quantities. This last stage is very difficult because this element is very stable in the soils. Willems and van Nieuwstadt (1996) have proved the difficulty of increasing the sward diversity after having stopped fertilization in the case of phosphorus-rich soils. Indeed, phosphorus is strongly linked to the soil (mainly to the clays, limestone and minerals such as iron and aluminium oxides) which explains the slow decrease of its content in time. Moreover, the quantities of this element exported in forages are very small (10-20 kg.ha^{-1}.year^{-1}) (Marrs, 1993).

As shown in figure 3, the number of species decreases also when the potassium content is over 30 mg/100 g of dry soil. However, the maximum species number is observed when the content (about 20 mg/100 g) corresponds to an optimal plant nutrition in this element (Commission des sols de Wallonie, 1988). This reference must be considered with caution since it is obtained from routine laboratory. We can nevertheless assume that moderately high potassium contents are compatible with high values of the species number. This opposes this element to the extractable phosphorus for which this maximum diversity is much lower than the contents which are considered as optimum for productive agricultural lands (5-8 mg/100g). Moreover, the content of this element decreases quite quickly if it is no longer applied because it is more leachable than phosphorus. Potassium is also exported by plants in greater quantities (about 200 kg.ha^{-1}.year^{-1}).

Other factors, as mentioned above, are known to modify the flora, such as nitrogen and the cutting regimes. The latters do not however constitute unavoidable obstacles to the diversity reconstruction. Indeed, their effect stops as soon as they are no longer applied. It must only kept in mind that intermediate regimes (2 cuts per year) are in general more compatible with species diversity in herbaceous swards. Nitrogen is a particular case because the available nitrogen, mainly nitrate, is very labile and quickly leaches in the soils, but it can be renewed by different sources. For this reason, its pool is very difficult to measure.

The first source, which can hardly be avoided, concerns the atmospheric deposits which, in some regions, can be very high (50 kg/ha in the Netherlands) (Willems et al., 1993).

The second one concerns the legumes ability to fix atmospheric nitrogen by symbiotic associations.

The third one comes from the mineralization of the organic matter which, in certain soils, releases high quantities of nutrients which offer the possibility of maintaining constant productivity levels during several years after the cessation of fertilization (Berendse et al., 1992). In the case of wet grasslands, this factor could be controlled by an adequate management of the water table (Oomes et al., 1996). The mineralization of the organic matter can be prevented in this case by anaerobic conditions. The removal of the top soil is another method, undoubtedly difficult to be applied to large areas but radical in its effect.

When the nitrogen inputs (by fertilizers or atmospheric deposits) are insignificant, the available soil phosphorus could intervene in the control of nitrogen availability and then on the plant diversity. According to this hypothesis, the available nitrogen would remain the most important factor limiting the plant diversity; its effect occuring indirectly through the advantage that it offers to productive species. The latter have, indeed, the ability to replace the other species through their higher competitivity, not only at root levels but also at the aerial part levels. This competitivity occurs through high and rapid biomass production when the nutrient level is sufficient. The other species are rapidly in the shade of the other ones and disappear.

<u>A first hypothesis</u> of the indirect effect of the soil available phosphorus would be its control of the legume content of the sward. It has been stated for quite a long time that, contrarily to the nitrogen supply (Willems et al., 1993), the phosphorus supply to a deficient

soil highly increases the proportion of legumes (Bobbink, 1991; Elisseou et al., 1995; Holford and Gleeson, 1976; Mamolos et al., 1995). This would greatly increase the nitrogen input by symbiotic fixation. Many studies, carried out specifically on *Trifolium repens L.* (Baines et al., 1983; Caradus and Snaydon, 1988; Holford and Gleeson, 1976), have shown the same tendency. Moreover, this element is necessary to nodulation (Dunlop and Hart, 1987; Marschner, 1995). Hart et al. (1981) and Hart (1982) have demonstrated that, when the phosphorus levels are small, the species dry matter yield was not modified by a nitrogen supply or a Rhizobium inoculation. This confirms that low levels of soil available phosphorus can limit the presence or the abundance of legumes in the sward. However, as far as our studied soils are concerned, this phenomenon could not be shown (figures 11 & 12).

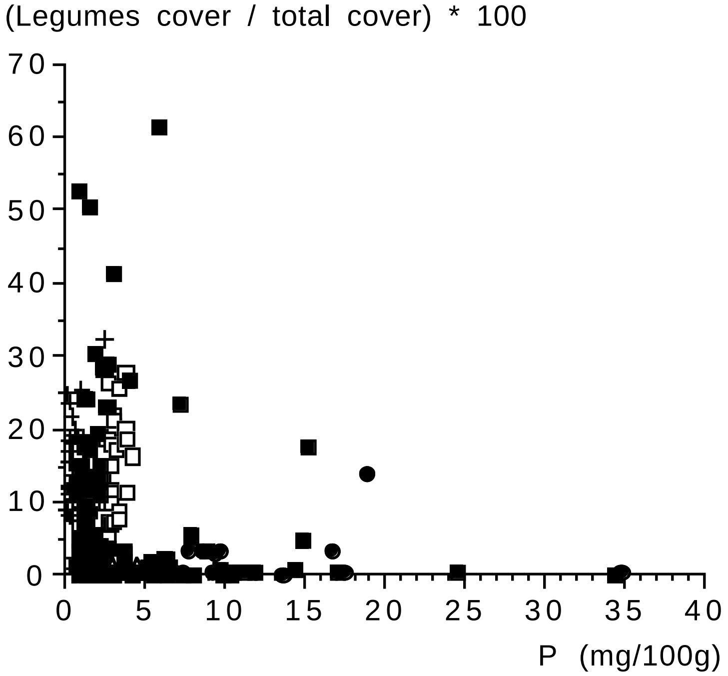

Figure 11. Relation between the soil available phosphorus and the percentage of legumes cover. (Legend : see figure 3)

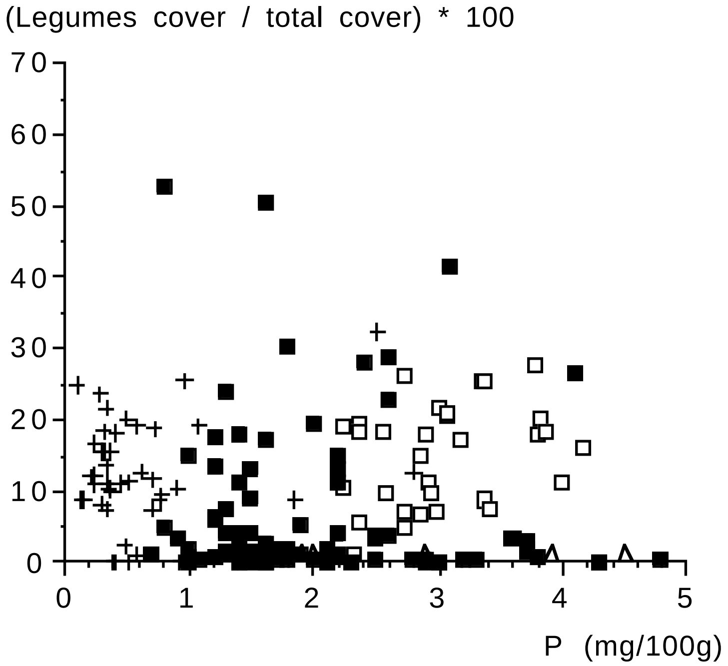

Figure 12. Relation between the soil available phosphorus and the percentage of legumes for plots where the available phosphorus contents are lower than 5 mg/100 g . (Legend : see figure 3)

Indeed, figure 11 shows that the highest relative legume covers (including essentially *Trifolium repens L.* and *Trifolium pratense L.*) can be related to low soil phosphorus contents. The legume cover decreases when exchangeable soil phosphorus exceeds 5 mg/100 g. This can be due to the fact that above this content, productive grasses are abundant and prevent the development of legumes. More intensive management systems and even, in some cases, higher quantities of available nitrogen could also play a role in the reduction of legume covers in the enriched soils. The latter could be explained by a second hypothesis, presented below. On the contrary, figure 12 does not show any relation between the soil available phosphorus contents lower than 5 mg/100 g and the legumes cover related to the total. This can be explained by the fact the limiting effect of phosphorus deficiency is not shown by a decrease in the cover of legumes. Nevertheless, if the legumes covers do not diminish in case of phosphorus deficiency, it is not obvious that the dry matter yields tend the same way. Nodulation and nitrogen fixation can be less efficient and yields can be affected. This absence of relation could also partly be explained by the fact that the phosphorus extraction method only partly reflects the real available phosphorus quantities in the poorest soils.

A second hypothesis concerns the limitation of soil organic matter mineralization by a deficiency of the soil available phosphorus (Amador and Jones, 1993; Marschner, 1995; Munevar and Wollum, 1977; Russel, 1973). Figure 13 shows that, if there is an effect of Nt on species diversity, a maximum species number is observed with soil Nt content between 0.4-0.9 %.

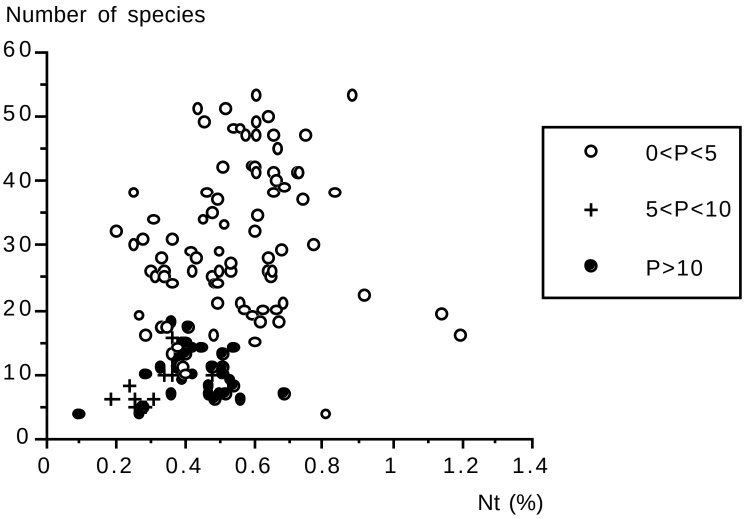

Figure 13. Relation between the total N and the number of species per 100 m^2 (the different symbols separate the soil exchangeable phosphorus levels (mg/100g dry soil)).

This maximum is reached only if exchangeable phosphorus content is less than 5 mg/100g dry soil. This could be explained by the fact that above 5 mg/100 dry soil, exchangeable phosphorus (directly or indirectly) increases the mineralization of organic matter and thus the production of N-NO3. The relative importance of these phenomena as well as their influence on the flora cannot be determined easily now. In order to obtain this precision, the available nitrogen contents should be assessed (mainly the nitric). However, the contents of this element in the soils are so small that it makes direct measurement very difficult on one hand and, on the other hand, indirect measurement by incubation does not exactly reproduce the real conditions. Another solution would be to analyse the plant nutrient content. This will be taken into consideration in a subsequent paper.

The first way through which the soil available phosphorus could limit the organic matter mineralization can be possibly the changes of composition (plants poorer in nutrients) or of the chemical structure (Cadish et al., 1994) of the plants which grow in these environments. Cheshire and Chapman (1996) have proved that plants (*Lolium perenne*), experiencing a phosphorus deficiency, showed high lignin contents. This could explain a decrease of the mineralization of the resulting organic matter and therefore a lower release of available nitrogen (Cadish et al., 1994). Another possibility, more direct, concerns the depression of microorganisms activity by a phosphorus deficient nutrition. This should concern more specifically the nitrfication processes. However, Hue and Adams (1984) have clearly demonstrated that nutritive solutions containing phosphorus concentrations of 0.004 ppm were enough to reach a maximum activity rate for Nitrosomonas and for Nitrobacter. This figure is below the values regularly obtained in soil solutions from species-rich grasslands (0.05 ppm; Janssens et al., 1996). However, attention should be paid to the fact that these two values, reflecting very different conditions, cannot be strictly compared.

As a conclusion, it could be assumed that the soil extractable P influences grassland biodiversity. This action could be indirect by its control on the soil available nitrogen quantities. This last element, according to this assumption, remains the main controlling factor of grassland diversity. It is then very likely that the phosphorus limiting effect is strongly reduced in arable land, poor in organic matter, where the N quantities mineralized each year are small anyway, whatever their phosphorus content can be. The influence of phosphorus on plant competition can also be direct. Finally, the soil micro-heterogeneities are extremely important as far as plant diversity is concerned (Gigon and Leutert, 1996; Willems et al., 1993). The interactions between available nitrogen and phosphorus and their influence on the vegetation probably interfere at this level through the relations described above and play an important role in the control of the grassland plant diversity.

VIII. RELATIONSHIP BETWEEN NUTRIENT CONTENT IN THE BIOMASS AND COMMUNITY DIVERSITY IN GRASSLAND

A. SUMMARY

Many studies have been carried out for several years in order to better understand the factors which influence the competition relationships between plant species in herbaceous swards. It is now clear that the dry matter production, when exceeding a certain threshold, limits the number of species which can coexist within a same community (Grime, 1979). On another hand, Janssens et al. (1997) have enlightened a similar effect of the soil available phosphorus.

This study concerns more specifically the nitrogen, phosphorus and potassium concentrations of the biomass sampled in 83 diversified grasslands. The nitrogen and phosphorus concentrations in the biomass did not show any significant relationship with the dry matter yields below 5 t DM/ha. This reveals a certain adaptability of the plants to produce a same quantity of dry matter from variable nitrogen and phosphorus assimilations. On the contrary, the depressive effect of the plants phosphorus contents on the plant diversity of the community is highly significant. This has not been observed for nitrogen. It is concluded that, when the soil available nitrogen is very low, the soil available phosphorus can play a direct role in the species competition, independently of the biomass production.

B. INTRODUCTION

It has been shown that the soil available phosphorus seems to be a factor which highly limits the grasslands diversity (Janssens et al., 1997). No community containing more than 20 species was found when the soil available phosphorus exceeds 5 mg/100 g. This depressive effect can be explained according to the Grime's theory (Grime, 1979). Indeed, according to him, the botanical diversity is strongly controlled by the community production (and so by the soil available nutrients like phosphorus); only a moderate production would enable the coexistence of a high number of species in the sward.

Grime (1979), Chapin (1980), Tilman (1990) and Marschner (1995) explain this trend by the species competition. The adaptations imposed to species by the nutrients-poor soils are such that only some oligotrophic ones can survive. An increase of the nutrient contents allows the installation of mesotrophic species and, in the same way, the increase of the botanical diversity. These mesotrophic grasslands are among the species richest communities in the temperate regions. Concurrent with the increase of the soils richness, new eutrophic species appear. These species adapted to rich soils grow quickly and use more nutrient resources than the other species which therefore overlap them (Elberse and Berendse, 1993; Mamolos et al., 1995). The oligotrophic or mesotrophics species are thus removed of the cover because of their smaller capacities of competition for light. The diversity decreases in such a way that eventually only some highly competitive species remain. This theory meets with the Tilman's hypothesis (Tilman, 1990) which differs from Grime's since the floristic relationships in poor soils are explained by the existence of an intensive competition for the nutrients; on the contrary, competition for light would be the main element explaining these relationships on rich soils.

The adaptability to soil nutrients deficiencies would express itself by a slow growth, a slow regrowth of leaves and a slackened reproduction. Tilman (1990) suggests besides that low nutrients contents in the biomass for a given production reflect this adaptability by showing a greater competition ability in these stress conditions. For these plants, when soil nutrient availability increases, the tissue concentration in these nutrients increases in a way to store resource for future less favourable conditions.

Our study aims to control this hypothesis by a study of the relationships between the biomass production and the nutrient contents of the plant communities and the diversity of these communities.

C. MATERIAL AND METHODS

The study has been carried out in Belgium (in Ardenne and in Gaume) and in Luxemburg.

Plots of series 1 (n=31) come from Gaume (named "Les Bulles"). The nutrient-poor soils are clayey with imperfect drainage causing great hydrological variations. Alliances contain the Caricion, the Mesobromion-erecti, the Arrhenatherion, the Molinion.

Plots of series 2 (n=43) have been selected in the Grand-Duchy of Luxemburg. Soils are wet, clayey, with a gley horizon, neutral to slightly acid and poor in nutrients. The alliances found contain : the Caricion, the Calthion (Senecio-Brometum racemosi and *Alopecurus pratensis* communities) and the *Arrhenatherion*.

Plots of series 3 (n=9) are located in the Bastogne area, in the Middle Ardenne. Soils are loamy-clayey, very slightly acid and contain nutrients-poor grasslands, either wet (*Filipendula* or *Comarum* community) or dry (Violo-Nardion).

Vegetation has been analysed by the Braun-Blanquet method (Braun-Blanquet, 1932) on 100 m^2 areas (10 m x 10 m). A rarity index has been calculated by using a relative rarity coefficient of each plant per region (Lambinon et al., 1992). The species have been attributed a rarity coefficient of 0 for the commonest and 5 for the rarest. Calculations have been carried out as follows :

Rarity index : $\sum_{i=1}^{s}\left(\frac{P_i*100}{\sum_{i=1}^{s}P_i}*Cr_i\right)$

The Shannon's diversity (Shannon, 1948) has also been calculated : $-\sum_{i=1}^{s}(P_i*\log_2 P_i)$

where : **P_i** is the proportion of the i^{st} species

Cr_i is the rarity coefficient related to the i° species

s is the number of species

The dry matter yields have been measured by cutting at 5 cm height in the beginning of July (1st decade) on 1 m^2 surfaces (in 4 repetitions for each plot). Samples (series 1 & 2) have been taken, dried at 105°C and analysed for total nitrogen (Kjeldahl) or mineralized in the oven at 450°C then solubilised in a nitric acid solution at 80°C. Phosphorus has been measured out by colorimetry and potassium by spectrophotometric analysis.

D. **RESULTS**

The relationships between the swards dry matter yields and their number of species per 100 m^2 or the Shannon index shows a similar curve to Grime's humped curve (Grime, 1979) (figures 1 & 2).

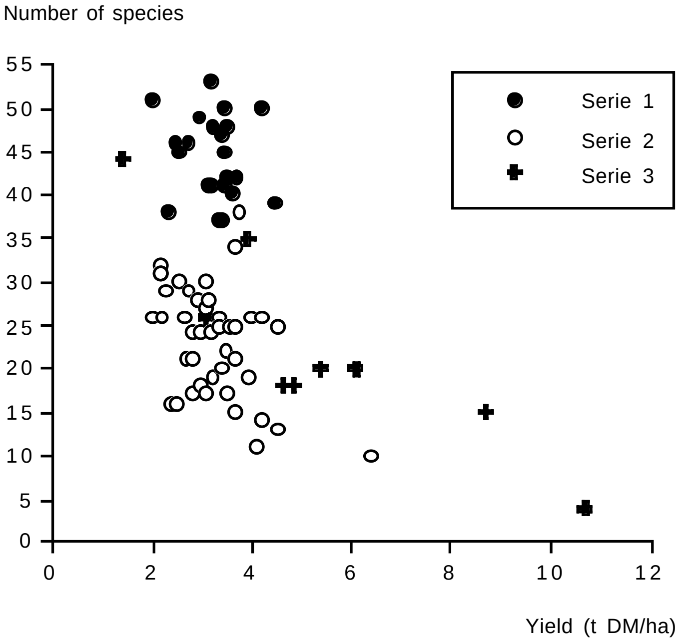

Figure 1. Relationship between the dry matter yield of the first cut (beginning of July) and the number of species per 100 m2.

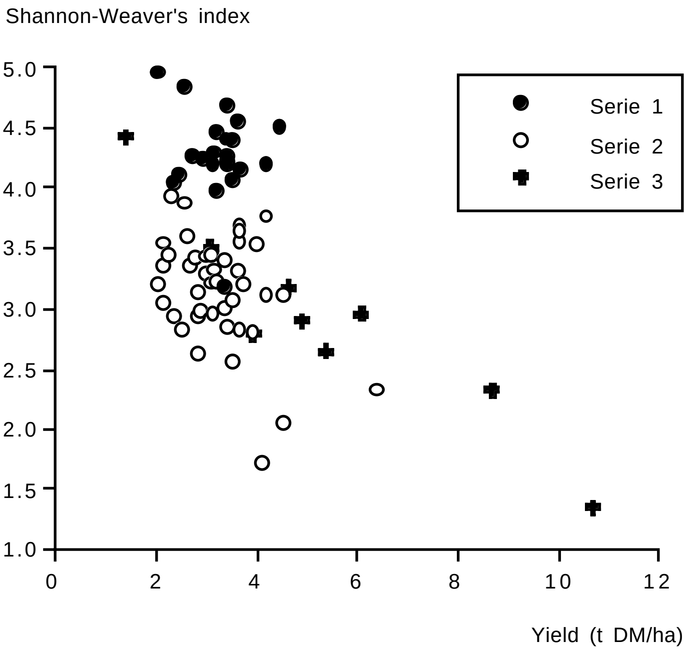

Figure 2. Relationship between the dry matter yield of the first cut (beginning of July) and the Shannon's diversity.

The nitrogen contents in the biomass are not linearly or logarithmically related with the diversity indices (number of species, Shannon index and rarity index) (table 1; figure 3).

Contents	N (% DM)	P (mg/100g DM)		K (mg/100g DM)	
Number of species	R^2=0.00	y=-47.8 log(x)+137.6	R^2=0.53*	y=-33.3 log(x)+135.4	R^2=0.17*
Shannon index	R^2=0.00	y=-2.7 log(x)+9.7	R^2=0.51*	y=-2.0 log(x)+9.8	R^2=0.18*
Rarity index	R^2=0.00	y=-1.8 log(x)+5.1	R^2=0.56*	y=-1.3 log(x)+5.1	R^2=0.19*

Table 1. Regression equations and determination coefficients between the indices used to estimate the botanical diversity (y) and the biomass N, P and K contents (x) (n=74) (indicate a high significant relation for a<0.001)*

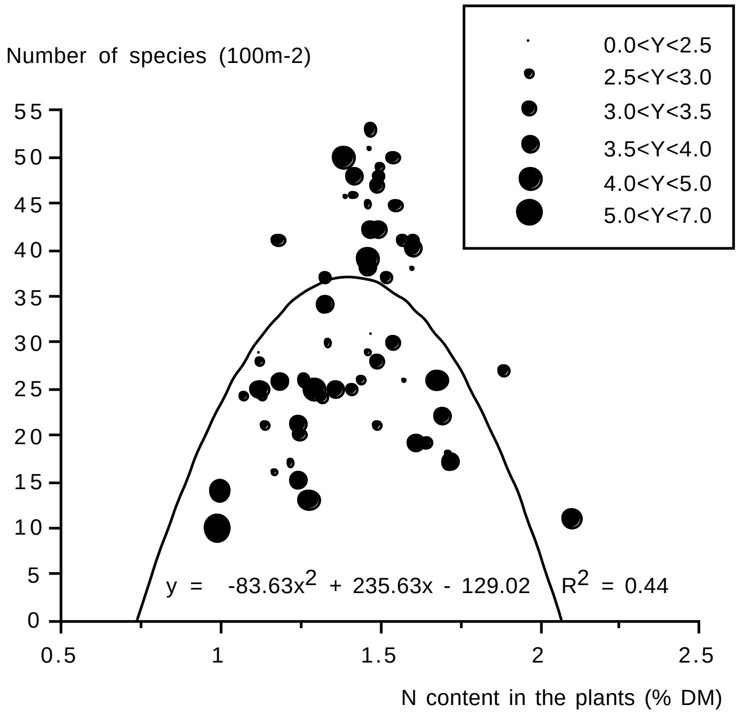

Figure 3. Relationship between the nitrogen concentration in the biomass and the number of species per 100 m^2 *(differentiated according to yields (Y) (t DM/ha))*

On the contrary, these three coefficients decrease significantly with the biomass phosphorus contents (figure 4).

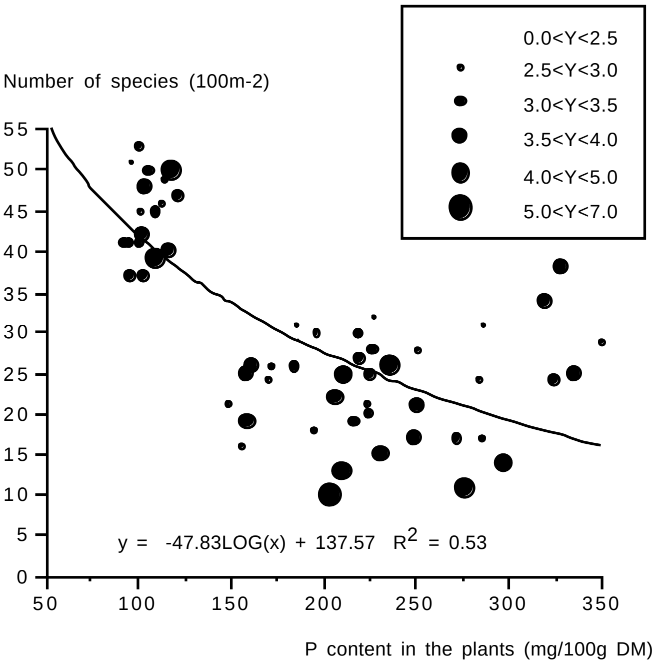

Figure 4. Relationship between the phosphorus concentration in the biomass and the number of species per 100 m^2 (differentiated according to yields (Y) (t DM/ha))

It should be noted that dry matter yields are not related to the biomass nitrogen and phosphorus contents (figures 5 & 6).

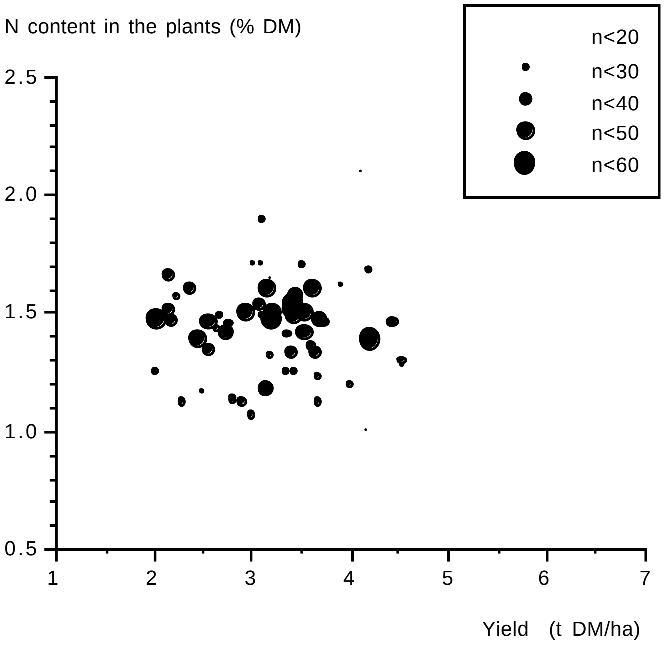

Figure 5. Relationship between the biomass nitrogen content and the DM yield obtained with the first cut achieved at the beginning of July. The different points size distinguish the number of species (n) per 100 m²

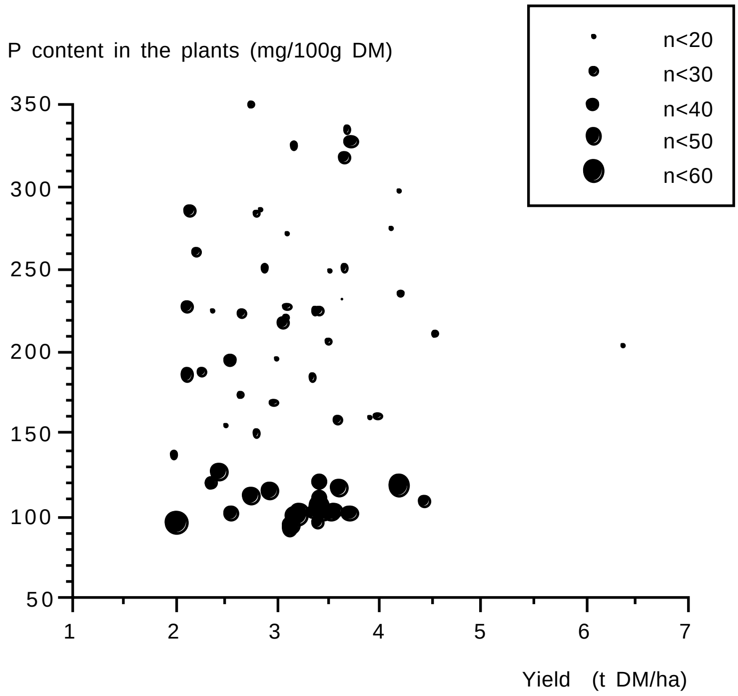

Figure 6. Relationship between the biomass phosphorus content and the DM yield obtained with the first cut achieved at the beginning of July. The different points size distinguish the number of species (n) per 100 m²

The utilisation of N, P, K yields does not increase significantly the relationships with the three indices used to assess the botanical diversity (table 2).

exports	N (kg/ha)	P (kg/ha)	K (kg/ha)
Number of species	R^2=0.00	y=-40.1 log(x) + 60.1 R^2=0.50*	y=-34.9 log(x) + 87.9 R^2=0.23*
Shannon index	R^2=0.00	y=-2.35 log(x) + 5.28 R^2=0.50*	y=-2.13 log(x) + 7.06 R^2=0.25*
Rarity index	R^2=0.00	y=-1.51 log(x) + 2.11 R^2=0.50*	y=-1.29 log(x) + 3.12 R^2=0.22*

Table 2 - Regression equations and correlation coefficients between the indices used to estimate the botanical diversity (y) and the N, P and K yields (x) (n=74) (indicate*

a high significant relation for $a<0.001$)

E. DISCUSSION

The relationship found between the dry matter yields and the number of species is similar to the humped curve, described in 1979 by Al-Mufti et al. and in 1979 by Grime, and reproduced afterwards by various authors (Wheeler and Giller, 1982; Wheeler and Shaw, 1991; Wilson and Shay, 1990). The maximum number of species per 100 m^2 vary between 2 and 5 t DM/ha. Over 5 t DM/ha, this number of species highly decreases until containing only maximum 20 species. This seems to support the hypothesis that competitive species exclude the others adapted to lower nutrient when the biomass production increases.

The essential elements as far as the botanical competition is concerned are nitrogen (Elisseou et al., 1995; Marrs, 1993; Mountford et al., 1993; Willems et al., 1993) and phosphorus (Elisseou et al., 1995; Janssens et al., 1997; Willems et al., 1993). Janssens et al. (1997) have enlightened the relationship existing between the number of species in a herbaceous sward and the soil available phosphorus. No grassland containing more than 20 species was found when the soil available phosphorus was exceeding 5 mg/100g (acetate-EDTA extraction according to Läkanen and Erviö, 1971). It seems thus that phosphorus, like nitrogen, plays an important role in the competition regulating the diversity of a herbaceous sward. As far as potassium is concerned, it does not constitute a limiting element since a maximum diversity is obtained with adequate potassium values for the plant nutrition (Janssens et al., 1997). Verhoeven et al. (1996) have also demonstrated the minor importance of potassium at this level.

The use of nutrient concentrations in the biomass of swards which have been sampled at the same development level seems to be an easy and reliable method to estimate the soil nutrient deficiencies limiting the biomass production (Koerselman and Meuleman, 1996; Verhoeven et al., 1996). This method could allow not to carry out analyses of available nutrients in the soil. Moreover available nitrogen is very difficult to measure because of its large variability with time.

Our results show a highly significant relationship between this species diversity and the biomass phosphorus contents (figure 4; table 1). This is not the case for the biomass nitrogen contents (figure 3) for which no linear or logarithmic relationship was found. However, figure 3 shows that a maximum number of species is observed for nitrogen contents amounting about 1.5 % DM. This is very likely explained by the fact that the P availability is also very low in these plots (less than 130 mg P/100 g DM). Indeed, swards with biomass containing 1.5 % nitrogen but more than 130 mg P/100 g DM, contain less species (figures 3 & 4). The potassium contents, despite their significant relationship with the botanical diversity, do not have to be taken into consideration for the above-mentioned reasons. Moreover, the determination coefficients between the biomass potassium contents or yields and the diversity are clearly lower than for phosphorus (tables 1 & 2).

It is thus likely that below 5 t DM production/ha, phosphorus can be an important element in the competition strategies of species characteristic of highly diversified plant communities. Meanwhile, in our sample, the dry matter yields below 5 t DM/ha, are not related with species diversity. In addition, these dry matter yields are not related to the biomass phosphorus (figure 6) concentrations. The influence of the biomass phosphorus contents on the herbaceous swards diversity cannot, in this case, be explained through the production of biomass. The competition between species would not be expressed, at this level, in terms of vegetal production but rather in terms of biomass nutrients concentration.

Species belonging to the most diversified communities show lower phosphorus contents (figure 6) for an equal dry matter production. This is compatible with Tilman (1990) and White (1972 & 1973). Chapin (1980) did also find this with plants growing in undisturbed natural habitat. This relation could not be reproduced for nitrogen (figure 5).

The absence of relation between the nitrogen or phosphorus contents and the dry matter production in the herbaceous swards indicates that species have different adaptations as far as the absorption and use of nutrients are concerned. These adaptations can be a higher relationship root/(stem+leaves). Some adaptations are specific to a nutrient : mycorrhiza associations or acid excretions by roots enable the plant to absorb less available phosphorus. The fixation of atmospheric nitrogen by legumes or the absorption of this element in the form of ammonium or amino acid constitute other demonstrations of the adaptation to the specific nitrogen absorption.

As a conclusion, the botanical diversity of the sampled grasslands is not strictly and exclusively related to the dry matter production. As far as these grasslands are concerned, no relationship can be stated between the indices used to assess species diversity and the dry matter production (below 5 t DM). These last indices are, nevertheless, related to biomass phosphorus concentration. The more diversified sites shows the lowest biomass phosphorus contents. This can be explained by the species adaptations to poor soils enabling them to produce the same dry matter quantity from different amounts of biomass phosphorus concentrations. This seems to indicate that when the soil nitrogen content is below a certain threshold which does not allow eutrophic species to grow, phosphorus might become the main element directly regulating plant competition. As soon as the nitrogen contents increase, eutrophic species can grow and exclude the other species.

This may explain difficulties faced in the attempts of restoring diversified herbaceous swards. Most of the surfaces which could be used to this end have been subject to considerable fertilizations due to the intensification of agricultural methods. The cessation of fertilization cannot often be sufficient to achieve a situation which could be favourable to the restoration or the reconstruction of diversified plant communities. In this case, the soil available nitrogen quickly decreases due to bare soils lixiviation or to vegetal exportation by cuts. Apart from atmospheric deposits, the renewal of these contents mainly depends on the sward legumes proportion and on the soil organic matter content. The soil available phosphorus could then play an indirect role by influencing these last two factors (Janssens et al., 1997). Under a certain level, to be determined, the importance of the soil available nitrogen in the control of the botanical competition could decrease in favour of phosphorus. This element concentration in the soil solution is very stable with high quantities linked to iron or aluminium oxides, to clays or to calcium. Indeed, these quantities can become available more or less quickly. In order to deplete the soil P contents, the low vegetal exports (10-20 kg/ha.year) of this element by cuttings are often insufficient. Radical (top soil removal) or specific methods (spreading of substances diminishing the available quantities of this element for the vegetation such as iron oxides, clays,...) have then to be used.

IX. ESTIMATION OF THE POTENTIAL PLANT DIVERSITY IN SWARDS BY QUICK METHODS

A. SUMMARY

The biodiverse swards becoming more and more scarce, a restoration or a reconstruction of this kind of vegetation is of major concern. This undertaking should be considered in an ecological network to prevent the isolation of the communities. Therefore, some lands (like road verges, industrial zonings, council or private garden, agricultural less productive parcels,...) could be used in this purpose. Meanwhile, many soils of these lands have high available nitrogen and phosphorus status. In previous works, the authors have shown the negative influence of a high trophic level of soil on botanical diversity in swards. In these works, it has been shown that species diversity in sward cannot exist above a certain limit of exchangeable phosphorus. Prior any attempt to restore species-rich sward, soil analyses should be done which is money and time consuming. Therefore, it would be very useful to have a method quicker, easier and cheaper allowing to assess the potentiality of restoration of such swards.

This research concerns the study of the validity and the practical interest of the N Ellenberg index in this context. Botanical assessments, according to the Braun-Blanquet method, and available soil nutrient analyses have been carried out on 273 grasslands sites. The grasslands have been chosen in many european region in a way to cover the largest range of vegetal communities.

The N Ellenberg index has been compared with other parameters corresponding to vegetal diversity (number of species or the rarity index). The N Ellenberg index was also confronted with soil and vegetal analyses. These studies allow a more precise comprehension of this index. Afterwards, the possibility of application of this method to predict the potential of restoration of species-rich sward on any site is discussed.

It then appears that this coefficient can inform about the potential diversity of species that may lie hidden in a grassland.

If the releves cannot be done by a botanical specialist, quicker and easier methods might

be used. The number of dicots species per 100 m^2 for instance could be proposed in this purpose.

B. INTRODUCTION

Since the application, these last decades, of intensive methods of agriculture in grasslands, the number or the surface of species-rich swards has dramatically decreased (Baldock, 1990). Only few biodiverse grasslands have persisted in marginal agricultural places or in natural reserves. These are isolated without any possibility of genes exchanges (Spellerberg, 1991). The purpose to restore such swards needs therefore their insertion within an ecological network. A large part of our landscape like road verges, industrial zonings, council or private gardens or agricultural marginal parcels can be used for the restoration of this ecological network. This use may lead into some difficulties regarding the development and the survival of these communities. These difficulties concern essentially the species supplies and the soil caracteristics. The first ones will not be considered in this paper.

Biomass production is known for many years to influence the botanical diversity of herbaceous swards (Grime, 1979; Wheeler & Giller, 1982; Vermeer & Berendse, 1983). Subsequently, many studies have shown that the soil available nitrogen level is an important factor limiting the botanical diversity in grasslands (Elisseou et al., 1995; Marrs, 1993; Tallowin et al., 1994; Willems et al., 1993). This limitation is assumed to be due to a higher biomass production. The botanical diversity of swards (estimated by the number of species, the rarity of the species or the Shannon index) is also linked to the soil exchangeable phosphorus as well as to the plant phosphorus contents (Janssens et al., 1997). These authors did never found species-rich swards with more than 20 species per 100 m^2 on soil with an exchangeable phosphorus status exceeding 5 mg/100 g (extraction : Lakanen & Erviö (1971); Cottenie et al. (1975)). This relation shows that soil analyses should be realized prior any species-rich swards restoration or reconstruction.

The authorities (for the road verges and the public parks), the nature protection organisms or even everybody who desire a diversified sward in his garden, need to know if their plans are realist. Agriculture is also concerned since the reform of the PAC in 1992 has evolved the agri-environmental program (CE 2078/92). This program concerns, among others, the species-rich grasslands. It is therefore imperative to assess a priori the potentiality of a site to support such a vegetation. Moreover, this estimation will have to be done on a very large scale if either the road verges, the public and private garden or the agri-environmental program are concerned. Therefore, the method has to be easy, cheap and quick. This method should for these reasons not implies soil analysis.

In order to estimate the nutrients limitations existing in diversified swards, recent studies (Boeye et al., 1997; Koerselman & Meuleman, 1996; Verhoeven et al., 1996; Wassen et al., 1995) have focused on the nutrient contents in vegetal rather than the nutrients in the soil. These works are mainly carried out in wet grasslands, and focus on the N and P contents in the vegetal tissues. It is assumed in these studies that the ratio N : P allow to know which nutriment limits the vegetal growth, which is related to species diversity. This method needs also chemical analyses. Moreover, the plants nitrogen contents vary very much with the physiological stage. Ideally, all the plants should be cut at the same physiological stage. For these reasons, this method, even if it is adapted for research for example, is not suitable in our purpose.

Ellenberg et al. (1991) have classified a great number of vegetal species according to different ecological variables such as soil available nitrogen. Each species is ranked in relation to its need in nitrogen. It is then possible to calculate a nitrogen index for a plot by adding the proportion of the covered surface by each species multiplied by its N Ellenberg coefficient. This nitrogen index could be useful to judge the nutrient status of a grassland without using a chemical analysis. These indices have been established for Central Europe. Thompson et al. (1993) have although shown that these can be used in England. Ellenberg indices have already been proved very useful and reliable in different conditions : for example to assess the risk of eutrophication in wetlands (Latour et al., 1994), the intensification of grasslands (Melman et al., 1988; Hill & Carey, 1997) or the soil quality (Hawkes et al., 1997).

Even if the last method is accurate, it implies a deep knowledge of the grassland species. In some circumstances, it would be more useful to propose a method that can be done by everybody. Indeed, it can be assumed that in many cases, it will be impossible to have a botanical specialist to do such studies, either for disponibilities reasons or because it is to expensive; the surface concerned could also be too large to be studied in this way. This quicker and easier method will obviously decrease the precision of the prediction. It is thus usefull to find methods that can be implemented quickly without the help of an expert and with a sufficient precision.

C. MATERIAL AND METHODS

The study has been carried out in Belgium and in Luxembourg. The sample includes 273 stations (figure 1) with a wide range of soils regarding their nutrients content, their pH, their texture and their water supply (table 1).

Series	Country	Location	Soil type	n
1	Belgium	"Viroin-Hermeton" Park overlapping Ardenne and Famenne region	Ardenne : heavy loam with a good natural drainage, or clayey soils with moderate to poor natural drainage or loamy to loamy-stony soils acid and very wet. Famenne : mainly calcareous and clayey and are well drained.	129
2	Belgium	"les Bulles" in Gaume region	clayey with imperfect drainage (great hydrological variations).	31
3	Belgium	Herve region	mainly loamy, wet, weakly acid and rich in nutrients (very intensive agriculture)	26
4	Belgium	Walloon Brabant	loamy and rich in nutrients (intensive agriculture)	13
5	Belgium	Bastogne area, located in the Middle-Ardenne.	loamy-clayey, slightly acid and relatively rich in nutrients	7
6	Belgium	Bastogne area, located in the Middle-Ardenne	like series 5 but chosen out of agricultural area, in wet (*Filipendula* or *Comarum* Community) or dry (Violo-Nardion) grasslands	8
7	Luxembourg	Gütland	wet, clayey soils, with a gley horizon, neutral to slightly acid	43
8 a	The Netherlands	Groningen (Brittany)	acid and wet moors (*Molinia caerulea* and *Erica tetralix*) (Bakker, 1989)	3
8 b	France	"la Poterie" near Lamballe	Dry moors (*Molinia caerulea* and *Erica cinerea*) (Géhu et al., 1988)	4
8 c	Belgium	Plateau des Tailles (Ardenne)	acid and wet moors (*Molinia caerulea* and *Erica tetralix*) (Dumont & Champluvier, 1989)	4

8 d	Belgium	Campine	acid and wet moors (*Molinia caerulea* and *Erica tetralix*) (Vanden Berghen, 1973)	5

Table 1. Description of the sites. (n = number of sites - total = 273)

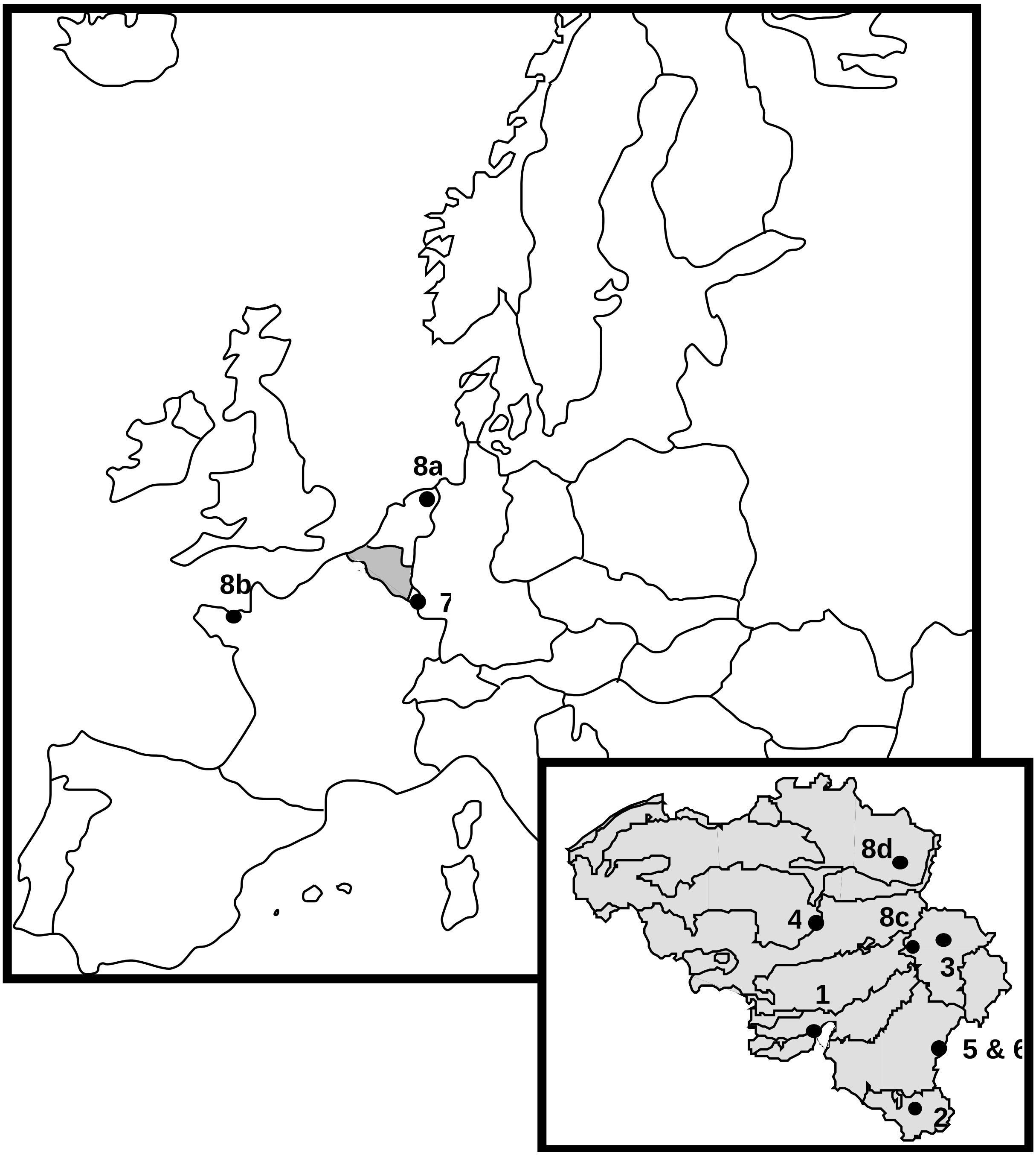

Figure 1. Localisation of the sites

All the stations are old permanent grasslands. Their management, very different from each other, have been applied for many years.

Vegetation has been analysed by the Braun-Blanquet method (Braun-Blanquet, 1932) on 100 m^2 areas (10m x 10m).

The vegetation releves have been subject to some mathematical conversions. A rarity index has been calculated by using a relative rarity index of each plant per region. This rarity index has been established by attributing a figure value to the appreciations of Lambinon et al. (1992) (from 0 = very common to 5 = very rare).

This rarity index has been calculated according to the cover, expressed in percentages, of each species (rarity weighted index).

$$\text{Rarity index : } \sum_{i=1}^{s}\left(\frac{P_i * 100}{\sum_{i=1}^{s} P_i} * Cr_i\right)$$

Where **P_i** is the proportion of the i^{st} species

Cr_i relative rarity index of the i^{st} species

s is the number of species

The N Ellenberg index has been estimated for each plot :

$$\text{N Ellenberg index: } \sum_{i=1}^{s}\left(\frac{P_i * 100}{\sum_{i=1}^{s} P_i} * N_{Ellenberg_i}\right)$$

Where **$N_{Ellenberg_i}$** is the N Ellenberg coefficient for the i^{st} species. (from 0 = species growing on soils very poor in nitrogen to 10 = species growing on soils very rich in nitrogen) (Ellenberg , 1991).

The dry matter yields (series 2 & 7) have been measured from cuts achieved at 5 cm height at the beginning of July on 1 m^2 surfaces (in 4 replications for each 100 m^2 station). Samples (250 g) have then been taken (series 2,3 & 7), dried at 105°C and analysed for total nitrogen (Kjeldahl) or mineralized in oven at 450°C then put at 80°C in a solution with nitric acid. Phosphorus has been measured by colorimetry. The soils have been sampled at a depth of 15 cm and analysed for exchangeable P (extraction by acetate-EDTA : Lakanen & Erviö (1971); Cottenie et al. (1975)) (table 2).

	Series 1	Series 2	Series 3	Series 4	Series 5	Series 6	Series 7	Series 8
Soil exchangeable P (mg/100g)	4.18 *5.87*	3.03 *0.55*	14.32 *3.75*	13.28 *7.65*	13.79 *3.44*	2.80 *1.18*	1.18 *0.21*	- -
Plant N content (%)	- -	1.47 *0.10*	3.28 *0.42*	- -	- -	- -	1.39 *0.26*	- -
Plant P content (mg/100g)	- -	106.58 *9.45*	418.12 *30.54*	- -	- -	- -	229.82 *54.46*	- -
DM yield (t/ha)	- -	3.23 *0.57*	- -	- -	- -	- -	3.24 *0.83*	- -
n	129	31	26	13	7	8	43	16

Table 2. Characterisation (mean and standard deviation (italic) and numbers of stations (n) studied in each series.

D. RESULTS

In our sample, the species diversity (assessed either by the number of species or the rarity index) is not related to the dry matter yield (table 3). This suggests that at least another parameter is controling this species diversity. The relation between the exchangeable phosphorus in the soil and the number of species or the rarity index is significant (table 3). The soil exchangeable P is not related to dry matter yield. This allow to assume that soil exchangeable phosphorus can be used as an indicator of species diverity with more precision than dry matter yield.

This soil parameter in only an estimation of the soil available phosphorus. The nutrient concentrations in the plant are, at this level, more precise but also more variable with time. The N and P concentrations in the vegetal tissues are also significantly correlated to the number of species and the rarity index.

Finally, the N Ellenberg indices are also significantly related to these last parameters (table 3).

	Number of species (100 m^{-2})		Rarity index	
Yield (t DM/ha)	$y=44.70*10^{-0.06x}$	$R^2=0.13$	$y=1.14*10^{-0.03x}$	$R^2=0.02$
Soil exchangeable P (mg/100 g)	$y=23.15*10^{-0.02x}$	$R^2=0.30$*	$y=0.73*10^{-0.05x}$	$R^2=0.33$*
plant N content (mg/100 g)	$y=36.51*10^{-0.16x}$	$R^2=0.53$*	$y=1.97*10^{-0.33x}$	$R^2=0.75$*
plant P content (mg/100 g)	$y=57.07*10^{0.01x}$	$R^2=0.67$*	$y=2.83*10^{-0.01x}$	$R^2=0.72$*
N Ellenberg index	$y=95.28*10^{-0.12x}$	$R^2=0.37$*	$y=5.01*10^{-0.17x}$	$R^2=0.60$*

Table 3. Regression equations and determination coefficients between the number of species (per 100 m^2) or the rarity index (y) and DM yield, soil exchangeable P, plant N & P contents and N Ellenberg index (x). (indicates a high significative relation for $\alpha < 0.001$).*

The N Ellenberg indices are not significantly linearly correlated to the dry matter yields. On the contrary, they are correlated to the soil exchangeable P and even more significantly to the plants N & P contents (table 4). The correlation with plant P content is higher than with plant N content.

	N Ellenberg index	
Yield (t DM/ha)	$y=0.17x + 3.67$	$R^2=0.04$
Soil exchangeable P (mg/100 g)	$y=0.12x + 4.53$	$R^2=0.38$*
Plant N content (%)	$y=0.95x + 3.44$	$R^2=0.61$*
Plant P content (mg/100 g)	$y=0.01x +2.55$	$R^2=0.74$*

Table 4. Regression equations and determination coefficients between the N Ellenberg index (y) and the DM yield, the soil exchangeable P and the plants N & P contents (x). (indicates a very high significative relation for $\alpha <0.001$).*

Figure 2 shows the relation between the number of species per 100 m^2 and the N Ellenberg index within a wider range of sites (n=273). In this range, we have considered the previous sites where the plant contents have been analysed (n=74) and other sites from different regions allowing the study a wider range of Ellenberg indices. These allow to observe a curve similar to the "humped-back" curve obtained by Al-Mufti et al. (1977) and Grime (1979). This is explained by the fact that the added sites are either too poor or too rich to have a great number of species.

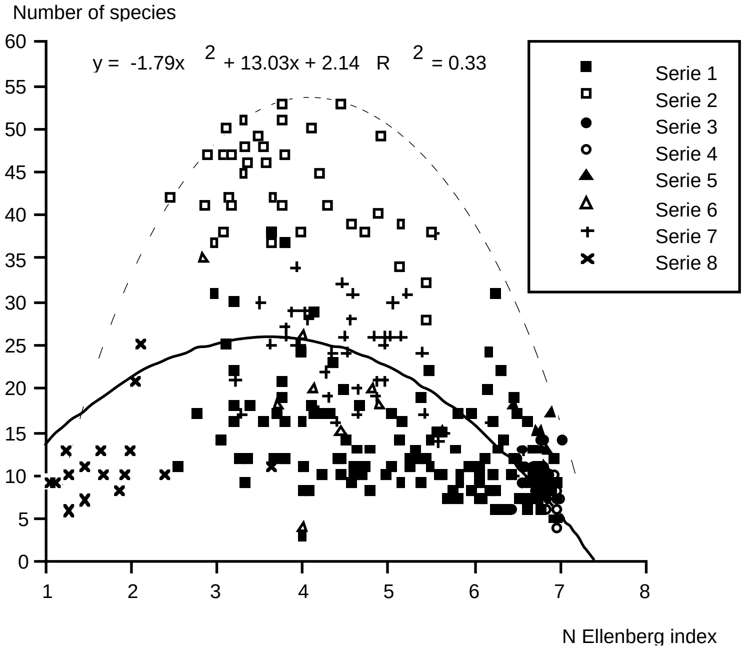

Figure 2. Evolution of the number of species with N Ellenberg index. The dashed line represents the envelope curve (the plain line represents the regression curve).

The decrease of the rarity index with the N Ellenberg index is highly significant (figure 3).

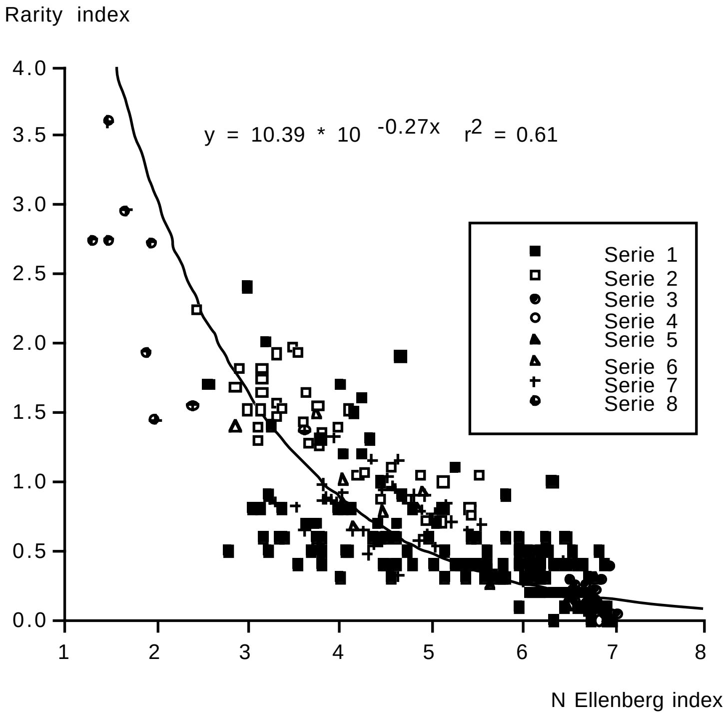

Figure 3. Evolution of the rarity index with N Ellenberg index.

Soil with exchangeable phosphorus status higher than 5 mg/100 g are always found with N Ellenberg indices above 5.5 (figure 4).

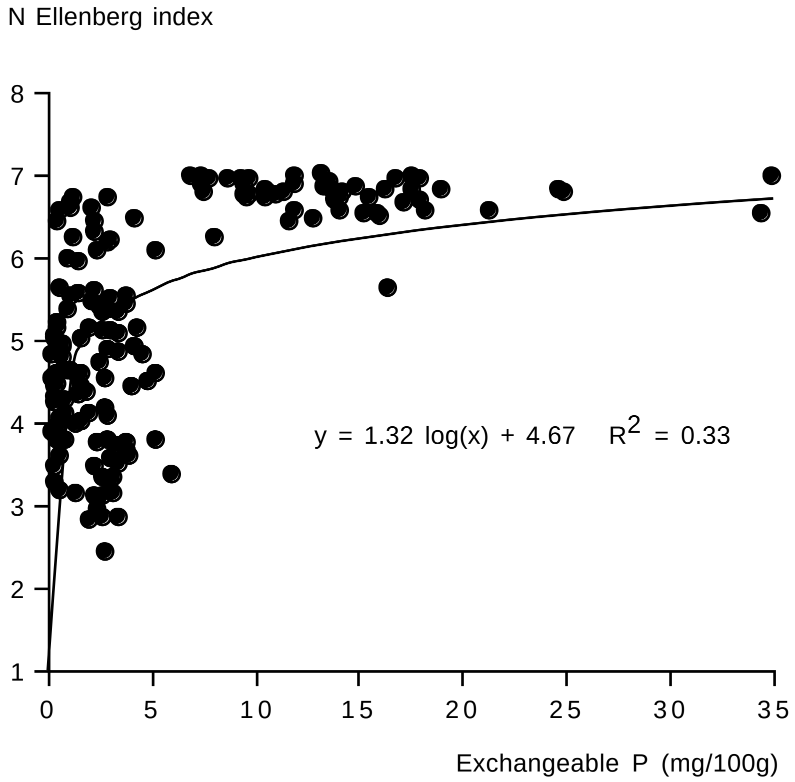

Figure 4. Evolution of the N Ellenberg index with the soil exchangeable P.

It could be useful to imaginate another method, that could be easier and that could be carried out without the help of a plant expert. In the next example, the number of dicot species is used to assess the soil exchangeable P status. Within our sites, no more than 10 dicots species per 100 m^2 were found on soils with exchangeable P higher than 5 mg/100 g (figure 5).

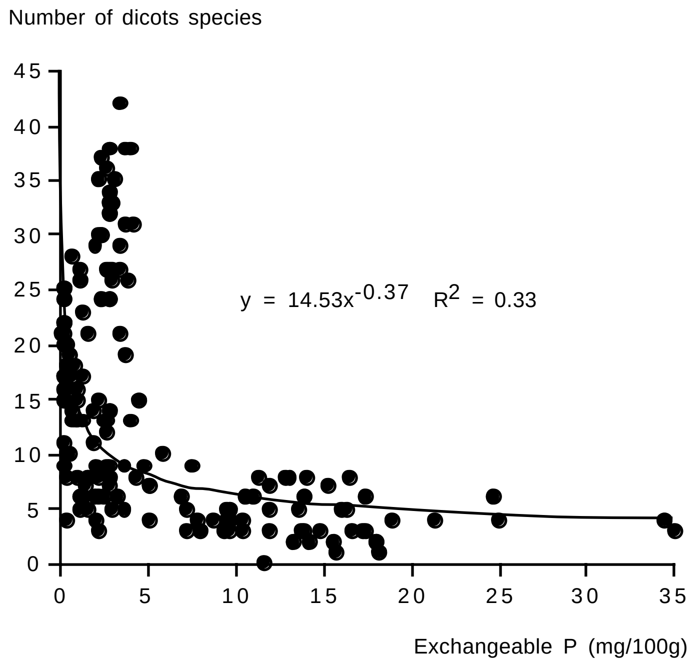

Figure 5. Evolution of the number of dicots species with the soil exchangeable P.

E. DISCUSSION

More and more people, individuals, public or private organisations are concerned or even involved in the restoration and the reconstruction of species-rich swards, including on road verges, in parks, ... In the purpose to assess the possibilities of success of these undertakings, it is necessary to define some parameters.

The botanical diversity of herbaceous swards is known for many years to be in relation with biomass production. Recent studies have gone further by enlightening the relation between this botanical diversity and the N and P content in plants. These elements are often cited as limiting the biomass production and influencing consequently the present flora (Chapin, 1980; Elberse & Berendse, 1993; Tilman, 1990).

In our sample, the dry matter yield is not correlated to species diversity (table 3). This is explained by the trophic level similarity of the chosen sites. This constatation does not refute the previous theory. Meanwhile, the fact that nutrient contents in plants in these samples are closely related to species diversity is interesting (table 3).

The same observation can be done when nutrient contents in plant are subsituted by soil exchangeable P (table 3). It is very difficult to assess soil available N by soil analyses; that is the reason why it has not been done in this study.

This indicates that plants of species-rich communities can have the same dry matter production as other communities with less soil exchangeable P and subsequently less N & P contents in their tissues. This implies physiological adaptations of these plants to produce the same dry matter quantities with less nutrients (Chapin, 1980; Elberse & Berendse, 1993; Tilman, 1990).

It appears therefore that a soil analysis could be a good clue, contrary to dry matter production, to indicate the potentiality of restoration of plant diversity. It has been shown, moreover, that there is no chance to see species-rich grasslands growing on soils with an exchangeable phosphorus status exceeding 5 mg/100 g; the others nutrient should not be considered (Janssens et al, 1997). Soil analyses could constitue a way of assessing the potentiality of survival of a species-rich community on a site. The time and the money involved in these analyses could be an obstacle to such a realisation. It must be kept in mind that this could be done by everybody and moreover sometimes on very large areas.

Another method regards the analysis of the nutrient contents of the plant. These are closely related to the vegetal diversity in our sample. Nevertheless, nutrient contents depend strongly on the physiological level of the plants harvested. A second factor influencing the nutrient contents of the plant is the specific potential of physiological or morphological adaptations (Grime, 1979; Mamolos et al., 1995; Tilman, 1990). It is thus necessary to sample all sites at the same period or at the same physiological stage of the plant. Moreover, the fact that it still needs a chemical analysis does also not favour this method.

It should be very useful to create an alternative method that could be reliable and easier.

The N Ellenberg index is, like N & P contents in the plant, significantly related to the vegetal diversity (number of species or rarity index) (table 5). When 247 sites are considered (figure 2), the envelope curve is similar to the "humped-back" curve obtained by Al-Mufti et al. (1977) and Grime (1979). It appears from table 6 that N Ellenberg indices are significantly related to plant P and N content (this is in relation with Meerts, 1997). When series 2 only is considered, plant P contents are significantly related to N Ellenberg indices (R^2=0.33; $p<0.001$) but not plant N content (R^2=0.001;NS). This suggest that N Ellenberg index does not explain only the N status of a site but rather indicates a kind of "nutritional index" of the studied community. This "nutritional index" will almost always be related to dry matter yield and this confirm the results found by Hill & Carey (1997). Moreover, this index is quite more precise since it allows to differenciate sites where N & P contents in plants are different but not dry matter production.

The "humped-back" enveloppe curve (figure 2) presents the plots where the botanical diversity is mainly explained by the variables taken into consideration by the N Ellenberg coefficients. The points located below this curve represent the plots where the diversity is limited by other factors such as cutting regime, species inputs (seeds rain, seeds bank),...

A maximum diversity is obtained for N Ellenberg indices about 4. The number of species decreases if these values are below 4. This can be due by a low nutrient status of the soil that only allows some adapted species to survive (Tilman, 1990; Grime, 1979). As a parallel, the diversity also decreases for N Ellenberg indices over 4. The competition for light should in this case be the reason of this decrease. Indeed, at these levels, the eutrophic species can express their greatest biomass production capacity and overlap the other ones (Tilman, 1990).

The use of this index would enable, in a first estimation, to avoid the use of the soil or vegetal analyses for estimating the possibility of restoration of species-rich swards. If, for example, one species impoverished sites (A) is considered (figure 6), with 20 species per 100 m^2 and a N Ellenberg indices of 4 then the limitation to increased botanical diversity (A') could be either because of inappropriate management and/or an impoverished soil seed bank or seed rain.

If two new sites are considered with the same number of species (20) but with N Ellenberg indices of 2 and 6 respectiveley (B & C). The first site (B) has a greater potential for improvement in its botanical diversity than the second (C). For both sites, the limitation to increased botanical diversity (A') could be still either because of inappropriate management and/or an impoverished soil seed bank or seed rain, but also because of nutrient availability. If nutrient availability is not changed, maximal diversity will be below 35 (B' & C') for both sites. For site B, the soil is too poor or have a particularity preventing high diversity (acidity, salinity,...). This particularity is often linked with a high rarity of the species (figure 3). The soil of site B should not be changed. The aim of conservation of this site should therefore focus on some species (rares, or of any ecological value). For site C, it can be assumed that the nutrient status of the soils has to be lowered before any important increase in species number can be expected. Figure 4 indicates that sites with exchangeable phosphorus exceeding 5 mg/100 g are found only with N Ellenberg indices over than 5.5.

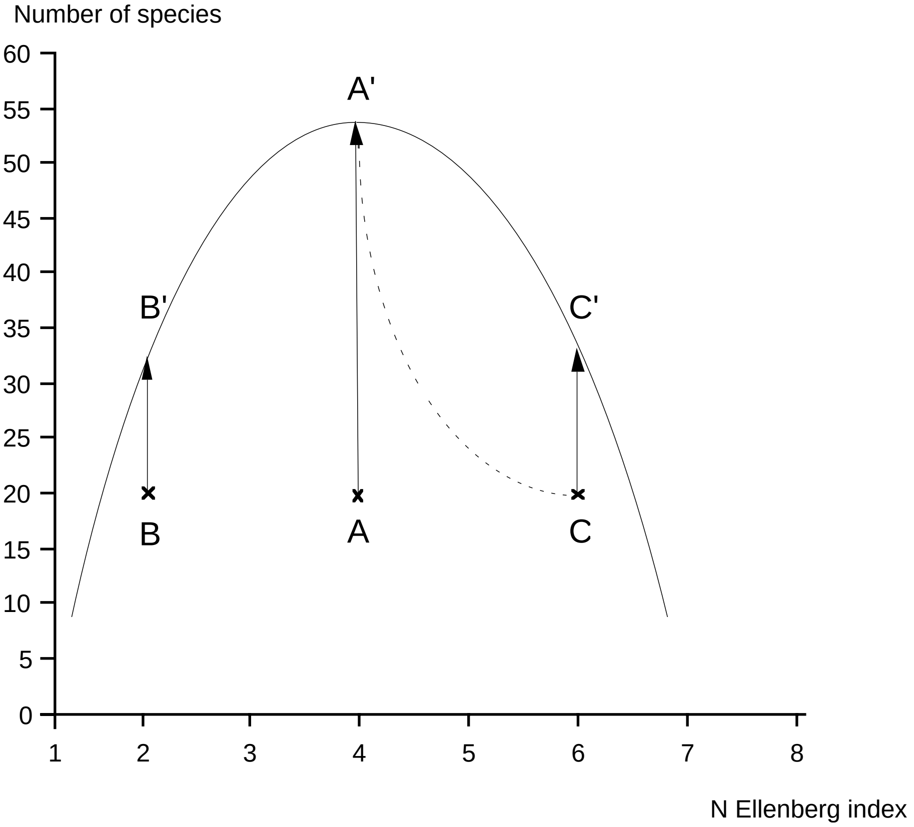

Figure 6 - Illustration of the N Ellenberg index use to estimate the restoration potentialities of the botanical diversity in the sward.

The rarity index significantly decreases with the N Ellenberg index (figure 3). This corresponds clearly to the explanation developed above regarding the "humped-back" curve. Indeed, on very poor soils, the only surviving species are strictly adapted to this environment. Since these environments are rare, these species are also. Concurrent with the enrichment of the environment, more and more species can live; these mesotrophic environments are notably less rare in Western Europe and in the studied area, particularly. On the richest sites, only some very common plants adapted to eutrophic environments survive, overlapping the others by their higher growing capacity. As a parallel with the number of species, above a N Ellenberg index of 5, the rarity index is lower than 0.5 (very common to common species).

It could be useful to use methods even more quicker which would be easier to implement and that would not require a great knowledge of the flora to estimate the potential botanical diversity of a plot. In the case of restoration of species-rich swards from already established swards, we assume that it could be possible to succeed in this restoration by an adapted management if the soil exchangeable P is low (below 5 mg/100 g) (Janssens et al. 1997). In this assumption we do not consider the species input (either by the seed bank/rain or sowing). A possible way to know if the soil exchangeable phosphorus status is greater or smaller than this limit could be a simple counting of the number of dicots. This can be done without identifying the species since it does not require a high botanical knowledge. It is also very easy to deduce from it the total number of species per 100 m^2 (in our sample : total number of species = 1.26 * number of dicots + 3.09 (R^2=0.96)).

In this case, more than 10 dicots by 100 m^2 have never been found with phosphorus status exceeding this limit (figure 5). This method need further work on different sites to be generalised and used. But it can be applied on large areas by everybody. That allows the restoration of species-rich swards to be more successfull by preventing the use of non suitable sites. But if this method is quicker and easier, it is also less precise. Indeed, if number of dicots is higher than 10, it might be assumed that exchangeable phosphorus is suitable for the restoration. Nevertheless, when number of dicots is lower than 10, it can be assumed that exchangeable phosphorus is above 5 mg/100 g but it is not undoubted. This low number of dicots may be due to an unsuitable management or an impoverished soil seed bank or rain. When quick methods are considered, this can only be enlightened by the N Ellenberg index method. All these methods need further studies but should constitute the base of prediction of restoration potential of a site when large area are concerned.

X. RESTORATION OF SOIL NUTRIENT CONTENT COMPATIBLE WITH BIODIVERSE SWARDS

A. SUMMARY

This study investigates the influence of nutrients on species-rich grasslands restoration and reconstruction. It was shown that low botanical diversity arised when high levels of soil elements, such as avalaible nitrogen or phosphorus, were encountered. Howewer, it should be borne in mind that, in order to recreate an ecological network, most areas likely to be reconverted into diversified grasslands are agricultural lands or road verges. These are often nitrogen- and phosphorus-rich soils. The first is often not a problem once it is not applied anymore thanks to its high mobility in soils, except in the case of high organic matter content combined with high mineralisation coefficient. Nevertheless, phosphorus is often the main factor limiting the botanical diversity in the herbaceous swards. Effectively, its influence on the botanical diversity combines with a low mobility in soils. Poor results are obtained from common methods aiming to deplenish the soil levels of this element.

Within this study, phosphate adsorption ability of iron oxides (industrial wastes) was investigated.

Firstly, tests were carried out with phosphate solutions. They enlightened the efficiency and kinetic of the reaction. Indeed, iron oxides adsorbed about 13 μmoles P/g oxide after one hour and 20 μmoles P/g oxide after one week.

Secondly, field experiments aimed to test the efficiency of iron oxides in soils. The presence of these compounds induced a great decrease in water soluble phosphorus. The decrease of the soil exchangeable phosphorus by the use of these oxides was not clear perhaps because of the extraction method (Läkanen & Ervio, 1971). Other treatments like soil removal or nitrogen-fertilized plant were also tested in their ability to decrease soil available phosphorus. Soil removal (20 cm depth) has very efficiently decreased soil exchangeable phosphorus. Nitrogen fertilized plant test has only decreased exchangeable potassium.

Finally, simple treatments, focusing on the specific area of iron oxides, showed a

dramatic increase in reactivity towards phosphate solutions. Thanks to these last treatments, the reactivity of the iron oxides was increased by 85 times.

B. INTRODUCTION

The restoration of species-rich herbaceous associations is an item becoming more and more studied in the ecological research undertaken during these last years. This trend arises from the great decrease of the areas covered by these associations. The main reason of this decrease resides in the intensification of agricultural practices in grasslands these last decades. Al-Mufti et al. (1977) and Grime (1979) have emphasized on the existence of a humped-back curve between the biomass productivity and the species density. This relationship shows a limited number of species adapted to conditions only involving a very low production. This very low production can be due to too poor nutrient quantities or to particular conditions affecting their disponibility (drought, salinity, acidity,...). Concurrent with the improvement of the conditions favourable to a higher biomass production, the number of species which can survive increases to reach a maximum. Over this maximum, more productive species, adapted to eutrophic conditions, appear. These species, thanks to a higher biomass production, compete with the other species which disappear because of a lack of light (Tilman, 1990). The sward botanical diversity decreases according to the dominance of these few productive species.

Janssens et al. (1997) have enlightened a comparable relationship between available phosphorus and the number of species per 100 m^2. In their work, it was shown that no station with more that 20 species per 100 m^2 was found when the soil exchangeable phosphorus (Lakanen & Erviö extraction) exceeds 5 mg/100 g. This element therefore seems, as well as nitrogen (Marrs, 1993; Willems, Peet & Bik, 1993; Tallowin et al., 1994; Elisseou, Veresoglou & Mamolos, 1995; Mountford, Lakhani & Kirkham, 1993), to be an essential soil nutrient influencing the botanical diversity. These two elements could interact under some conditions. Phosphorus could, in this case, influence the available nitrogen supply by controlling the organic matter mineralization or the fixation of atmospheric nitrogen by legumes. However, in the case where available nitrogen would be present in too small quantities, the available phosphorus could influence directly the botanical diversity (Janssens et al., 1997).

In view to optimalize the impact of the species-rich grasslands restoration at the landscape level, their insertion must be considered within the framework of the ecological network. The areas susceptible to fulfil this function mainly include agricultural lands and road verges. Because of storage of phosphorus in these soils, sometimes in great quantities, it seems obvious that the restoration of herbaceous diversified swards can be undertaken providing a decrease of these contents. This decrease of the soils available phosphorus content is rather difficult. Indeed, this element has the faculty of linking itself more or less strongly to other soils elements (calcium, iron and aluminium oxides, clays, ...) (Borggaard, 1983; Parfitt, 1989). The soils capacity to maintain the available phosphorus quantities for plants is often very high.

Different methods can be envisaged to decrease the soils available phosphorus content. The most efficient solution is to take out a soil superficial layer (Marrs, 1993). These layers are indeed the richest in nutrients and especially in phosphorus, thanks to its low mobility. Another solution, commonly used, lies in using a « vegetal pump » in order to extract soil nutrients. However, this method does not seem to be appropriate for phosphorus because this element is exported in very small quantities by the vegetation (10-20 kg/ha). In these conditions, for the greatest part of agricultural lands, several tens of years would be needed to impoverish the phosphorus reserves to an effective level (Johnston & Poulton, 1977; Marrs, 1993). A third method rely on the use of iron oxides. As natural component in soils, these elements fix phosphate ions and Modify the phosphorus distribution in the different pools of the soil. It could be assumed that these ions should be removed from the vegetal absorption. These different techniques have been tested in the Ardennes on classical agricultural lands; this study explains their limitations and potentialities.

Two experiments were carried out. The first one, in laboratory, concerns the iron oxides capabilities to adsorb phosphorus. The second was realised in the fields and compared the iron oxides with soil removal and plant exports.

C. MATERIAL AND METHODS

The iron oxides originate from a steel work factory of Cockerill in Belgium (Châtelineau). These oxides derive from industrial processes but can be considered as pure (table 1).

Element	**% (weight)**
Fe_2O_3	99.60
CaO	<0.02
SiO_2	<0.02
K_2O	<0.01
Al_2O_3	0.02
MgO	0.03
MnO	0.32

Structure : hematite.
Specific area : 5.8 m^2/g

Table 1. Total analysis of the original iron oxides (Bastin, 1996).

2.5 g of iron oxides have been mixed with 25 ml of a phosphorus solution (KH_2PO_4-200 ppm P). These mixtures have been shaken during one hour, one day, 1, 2 and 3 weeks. The analysis of non-adsorbed phosphorus has been carried out by colorimetric analysis (ammonium nitrovanadate; λ = 430 mm) of the resulting filtrate (filters « Gelman », 0.45 μm).

1. Field experiment

In 1993, an experiment was started on a rich arable soil near Bastogne in Belgium. This site was previously subject to intensive crop rotation (leys and cereals). The soil is a non-calcareous brown soil with clayey loam and high organic matter content.

4 replicates of 1.5 m x 5 m plots were settled for each treatment in a random complete block design.

The first test consists in a 0, 10 and 20 cm soil removal (plots 1, 2 and 3) made in June 1993. The second treatment tests the nutrient depletion by plant exportation.

The second test involved nitrogen-fertilized plants. Lolium multiflorum was used with 0 and 200 kg N/ha (Plots 4 and 5) cut twice a year (mid-July and September).

The iron oxides constitute the last test. 40 and 80 t/ha (plots 6 and 7) were spread mid-

June 1993.

The soils of tests 1 and 3 have directly been sown after treatment with species characteristics of the Alchemillo-Trisetetum association.

The first soil sampling was made in October 1993 (after one vegetation season). The second soil sampling occured in October 1995. The soils of each site have been sampled at a depth of 15 cm, dried (40°C), ground at 2 mm and analysed for : pH(H2O), organic matter (O.M.) (oxydable carbon : Walkley and Black 1934), total nitrogen (Kjeldahl) (Nt), exchangeable nutrients : P, K, Ca and Mg (extraction by acetate-EDTA : Läkanen & Erviö (1971); Cottenie, Kiekens & Verloo (1975); measured with atomic absorption spectrophotometer except for phosphorus (colorimeter)). The unit used to present the soil nutrient is mg/100 g dry soil. The organic matter and Nt contents are expressed as % dry soil. In 1995, only P, K and O.M. analyses were repeated.

Another soil sampling have been made 15 days after the spreading of the iron oxides to assess the water soluble phosphorus (soil/solution ratio: 1/10).

2. Improvement of the oxides fixation capacity

a) Miscellaneous treatments

The specific surface seems to be the limiting factor of the iron oxides reactivity towards phosphorus (Borggaard et al., 1990). The next stage of this experiment then consists in increasing this specific area by applying different treatments.

- 1-Blanco

pH fixation: 50 g iron oxides are suspended in 450 ml demineralized water. The pH of the suspension is stabilised to pH 6 with KOH or HCl (1N) and filtered.

Drying : the filter containing the filtration residue is placed into a stove (70°C; maximum ventilation).

Final rinse : the obtained oxide is then crushed and suspended in a solution of 300 ml demineralized water and is shaken during 2 minutes. The suspension is then centrifuged during 10 minutes at 2000 rpm. The supernatant conductivity is measured and the operation is started again until obtaining a constant value. The suspension is eventually filtered and the sample is drought in a stove (25°C).

- 2-Heating

50 g of iron oxides are heated at 900 °C during 2 hours. These oxides are then air-cooled. The various stages applied to blanco (pH adjustment, drying and final rinse) are then achieved.

- 3-Acid attack

50 g iron oxides are suspended in 300 ml H_2SO_4 (4N); the suspension is shaken at 25°C during 7 days and is then filtered. The filtration residue is again suspended in demineralized water and neutralised at pH 6 with KOH (1N). After filtration, the different stages applied to blanco (pH adjustment, drying and final rinse) are then achieved.

- 4-Simple precipitation

50 g iron oxides are suspended in 300 ml H_2SO_4 (4N); the suspension is shaken at 25°C during 7 days and is then filtered. The filtrate is collected. An iron oxide gel is precipitated afterwards by adding KOH (5N) drop by drop, until pH 12.4, to the filtrate which is continuously shaked by a magnetic bar. The obtained gel is filtered after having rested during 3 days at 25°C; it is then washed 5 times with 50 ml demineralized water. After filtration, the different stages applied to blanco (pH adjustment, drying and final rinse) are then achieved.

- 5-Combined precipitation

50 g iron oxides are suspended in 300 ml H_2SO_4 (4N); the suspension is shaken at 25°C during 7 days. No filtration is achieved at this stage. An iron oxide gel is precipitated by adding KOH (5N) drop by drop, until pH 12.4, to the filtrate which is continuously shaked by a magnetic bar. The obtained gel is filtered after having rested during 3 days at 25°C; it is then washed 5 times with 50 ml demineralized water. After filtration, the different stages applied to blanco (pH adjustment, drying and final rinse) are then achieved.

b) Specific area

The specific area is calculated (Bastin, 1996) with the BET method from an adsorption or desorption isotherm of a gaz mixture (30% N_2 and 70% He) on the oxide.

c) Phosphate adsorption

0.1 and 0.5 g iron oxides are suspended in 50 ml phosphate solution (KH_2PO_4 - 200 ppm P). The suspensions are shaken during 8 days and are filtered afterwards (filters « Gelman », 0.45 μm). Two repetitions are achieved. Phosphorus quantities in solution are measured with a colorimeter.

Only the results of the experiment involving 0.5 g iron oxide will be presented, except for the precipitation experiments (0.1 g iron oxide). This is justified by the fact that 0.5 g iron oxides treated by the simple or combined precipitation directly and totally fixed all the phosphorus present in the solution. These oxides were then not saturated, their maximum sorption capacity could then be deducted only by an experiment involving a weaker ratio (iron oxides quantities / phosphorus quantity).

D. RESULTS

1. Preliminary experiment

The preliminary analysis has enlightened the fast phosphorus adsorption on the iron oxides tested (fig. 1).

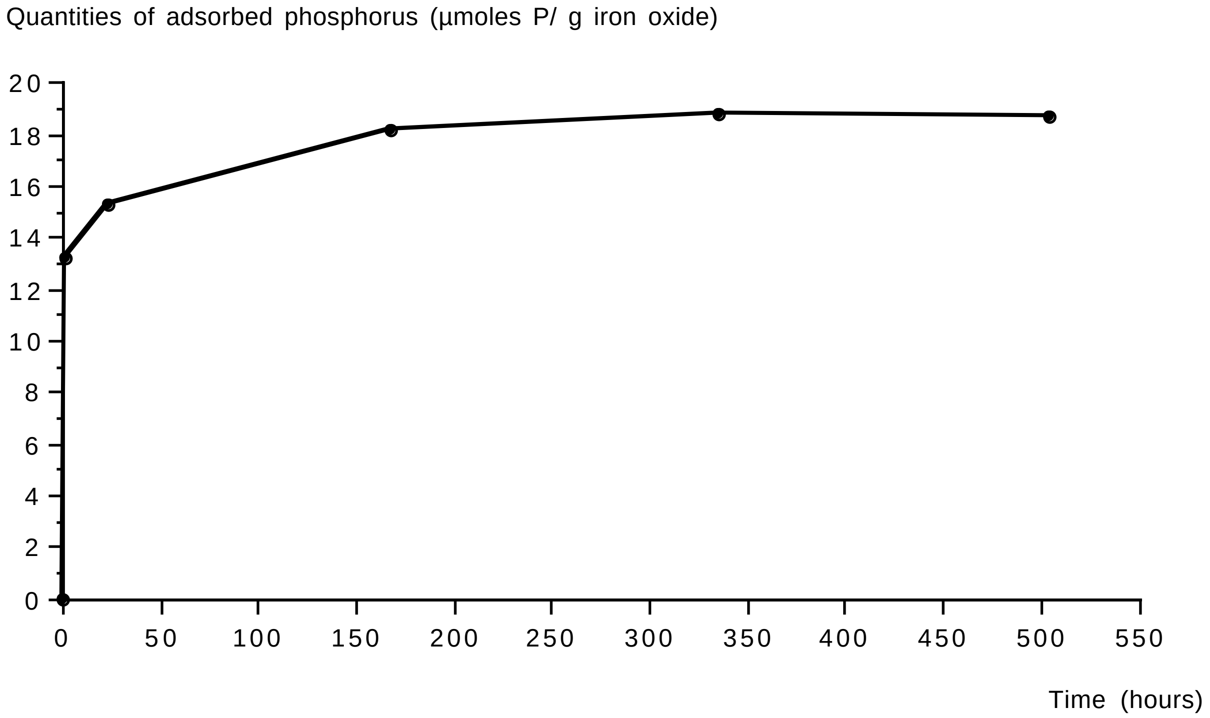

Figure 1. Quantities of adsorbed phosphorus on non treated iron oxides.

The oxides quickly decrease the concentrations of 30% of these initially present (200 ppm) in solution (this result is already reached after an hour and remains almost stable after 7 days). This result corresponds to a bit less than 20 µmoles P/g oxide.

2. Field experiment

The depressive effect of soil removal on the organic matter contents, exchangeable phosphorus and calcium, is significant independantly of the sampling date (table 2). The exchangeable potassium contents, even if they seem to decrease with the depth, are not significantly affected by this treatment at any date.

The use of « vegetal pumps » has only affected the exchangeable potassium contents. The iron oxides used have not diminished the exchangeable quantities of any of the analysed elements (table 2).

	M.O.	***s.d.***		**P**	***s.d.***		**K**	***s.d.***		**pH**	***s.d.***		***Ca***	***s.d.***	
1 (ref.)	4.0	*0.6*	**a**	3.2	*0.2*	**ab**	39	*8*	**a**	6.2	*0.1*	**a**	*170*	*58*	**ab**
2	3.5	*0.6*	**a**	3.0	*0.9*	**abc**	32	*3*	**a**	6.4	*0.1*	**a**	*135*	*18*	**abc**
3	2.2	*0.6*	**b**	0.9	*0.3*	**bc**	25	*3*	**a**	6.4	*0.1*	**a**	*88*	*10*	**bc**
1 (ref.)	4.0	*0.6*	**a**	3.2	*0.2*	**ab**	39	*8*	**a**	6.2	*0.1*	**a**	*170*	*58*	**ab**
6				3.7	*0.7*	**ab**	34	*4*	**a**	6.3	*0.1*	**a**	*145*	*14*	**ab**
7				2.9	*1.1*	**ab**	35	*4*	**a**	6.2	*0.2*	**a**	*115*	*19*	**ab**

1993 sampling

	M.O	***s.d.***		**P**	***s.d.***		**K**	***s.d.***	
1 (ref.)	3.8	*0.3*	**a**	2.6	*0.3*	**a**	18.9	*3.8*	**a**
3	2.4	*0.4*	**b**	0.5	*0.2*	**b**	17.2	*1.9*	**a**
4 (ref.)	3.9	*0.7*	**a**	2.7	*0.5*	**a**	16.9	*3.0*	**a**
5	3.7	*0.4*	**a**	2.7	*0.5*	**a**	9.9	*2.4*	**b**
1 (ref.)	3.8	*0.3*	**a**	2.6	*0.3*	**a**	18.9	*3.8*	**a**
7				2.6	*0.5*	**a**	20.8	*2.1*	**a**

1995 sampling

Legend :

1 : soil removal 0 cm

2 : soil removal 10 cm

3 : soil removal 20 cm

4 : Lolium multiflorum without nitrogen fertilization

5 : Lolium multiflorum + 200 kg/ha Nitrogen

6 : ferric oxide 40 t/ha

7 : ferric oxide 80 t/ha

Table 2. Results of the soil analyses (1993 and 1995 sampling). Comparison of the different treatments. The units are mg /100 g except for M.O. (%).

(The letters distinguish the groups for a Tukey test (0,05 %; n=4)).

However, the phosphorus analysis in the soil solution shows an important depressive effect of iron oxides. Fig. 2 shows that 80 tons iron oxides spread per ha decrease the phosphorus quantities in the soil solution to the level close of these commonly found in the species-rich grasslands soils (dotted line).

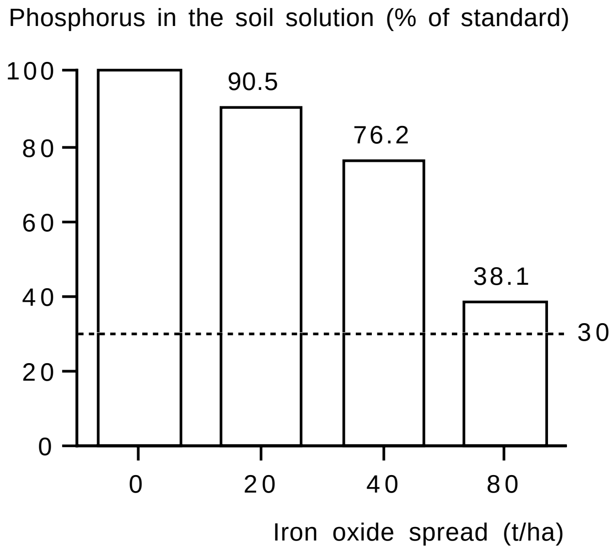

Figure. 2. Water soluble phosphorus in relation with iron oxide spread (t/ha). The results are presented in percentage (100% = 0 t/ha iron oxide (0.21 mg/l soil solution)).

The dashed line shows the level found in the species-rich grasslands of the region.

3. Improvement of the oxides fixation capacity

a) Specific area

The specific areas analysis of iron oxides obtained from the different treatments has shown a clear increase for the precipitation experiments and especially for the simple precipitation (table 3). The heating and acid attack have not increased the specific area.

Treatment	Specific area
Blanco	6.7
Heating	2.5

Acid attack	7.1
Simple precipitation	192.3
Combined precipitation	33.3

Table 3. Specific area (m^2/g) of the iron oxides resulting from different treatments.

b) Phosphates adsorption

Concurrently, the sorption capacity of iron oxides resulting from the different oxides treatments shows a high increase with the simple precipitation (fig. 3).

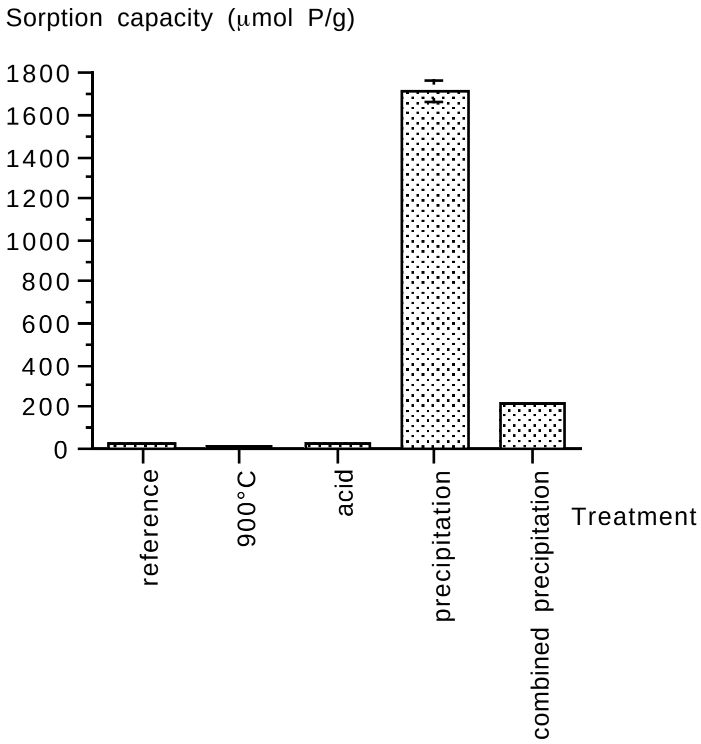

Figure 3. Sorption capacity (µmoles P/ g iron oxide) of the iron oxides resulting from the different treatments (the bars represent the standard deviation (n=2)).

This parameter has not been increased by heating and acid attack. This parameter seems to evolve in a linear way with the specific area ($y = 9.13 x - 47.27$; $R^2 = 1.00$).

E. DISCUSSION

The preliminary experiment in solution has enlightened the fast action of iron oxides. 13 μmoles P/g oxide has been adsorbed after 1 hour. The maximum adsorption (20 μmoles P/g) is reached after 1 week (fig. 1). This value, related to the oxid specific area, equals to 3.4 μmoles/m^2. This is consistent with the values quoted in previous studies (0.8 to 4.1 μmoles/m^2 (Torrent, Barron & Schwertmann, 1990); 1.5 to 6.9 μmoles/m^2 (Barron, Herruzo & Torrent, 1988). Only the order of magnitude can be taken into account because the conditions in which they have been obtained as well as the crystalline form of the oxides used can influence them strongly. This crystallinity degree is known to strongly influence the adsorption reaction via the specific area (Childs, 1992). According to Parfitt (1989), the phosphate sorption capacity evolves following this order: hematite < goethite < ferrihydrite; this order also corresponds to growing specific areas. The specific area of the initially used oxide (hematite) could be theoretically increased; the reactivity of this product should be therefore automatically improved. To this end, iron oxides have been submitted to different treatments (acid treatment with or without neutralisation, heating) in order to increase their reactivity.

Heating was not found to be an interesting treatment. This can be explained by change in structure which occured within the oxide due to the evaporation of the water present in the free spaces. Heating would then lead, according to this assumption, to a collapse of the wall of these free spaces and would therefore decrease the specific area and the reactivity of the product.

The acid attack did not show any positive results. The increase of the specific area and of the phosphates sorption capacity is not significant. The iron oxides quantities dissolved are much lower than those calculated theoretically (calculation based on the hematite solubility product at a certain pH (Schwertmann & Taylor, 1989)). According to Bastin (1996), the low dissolution obtained would be due to a kinetic problem essentially explained by a too short reaction time and temperature.

During the third treatment, the simple precipitation, the neutralisation of the dissolved iron oxides filtered solution caused the precipitation of a new amorphous component. This product was identified as ferrihydrite (Bastin, 1996). The specific area of this product (192.3

m^2/g) is 30 times higher than the specific area of the original product; this meets the values cited by Childs (1992) which lay between 200 and 500 m^2/g. The reactivity of this product follows the same trend since it is 85 times higher than the reactivity of the original product.

The combined precipitation is very comparable with the previous one except that the new product is mixed with the original hematite, much less reactive, consequently decreasing the performance of the product. According to Bastin (1996), the lower efficiency could arise, in addition to the dilution, from the precipitation of the product, neoformed on the hematite particles already present. The two stages would then lose a part of their common surface which would become unusable for adsorption.

The fixation potentialities of these products offer applications which could be related to other disciplines than those previously envisaged. The depollution of eutrophised waters, more and more debated, can constitute an application subject for these oxides. This requests however preliminary studies regarding the stability of products and links as well as the regeneration possibilities.

The removal of a superficial layer of soil has logically decreased the quantities of exchangeable elements; potassium remains an exception. The fact that the decrease of its contents is not significant can be probably explained by its mobility in soils. Within the framework of our objective aiming at the reconstruction of species-rich swards, this treatment is therefore appropriate since it diminishes the soils exchangeable phosphorus contents but not exchangeable potassium. However a major drawback of this treatment is its practical aspect which strongly limits its use. It can be applied in certain particular cases (natural reserves, didactic spaces, small gardens,...) on small surfaces.

The use of « vegetal pumps » receiving nitrogen has enabled to obtain inverse results to the previous treatment. Indeed, only soil exchangeable potassium, in our experiment, is affected by this method. This is mainly caused by greater potassium exports by the vegetation (approximately 300 kg/ha) than for phosphorus (20-30 kg/ha).

None of the described methods seem sufficient or applicable to diminish the available phosphorus contents because of the reasons cited above. Large-scale application of soil removal is not feasible, even if it is a very effective option. Exportation by vegetals requires a too long period of time. The low efficiency of this method towards phosphorus contents is related to the conclusions of Johnston & Poulton (1977) and of Marrs (1993) who state that several tens of years are generally requested in order to deplete sufficiently its contents. Another method preventing the assimilation of phosphorus by vegetation must then be found out. The use of iron oxides is fully justified since these elements are naturally present in the soils in which they are known for their phosphorus fixation properties. Besides, as great amounts of these are present in the soils, an additional supply would just slightly disturb their properties. Finally, iron oxides investigated in this study result from industrial processes; they display two more advantages : being available in great quantities while being pure.

The spreading of iron oxides has shown inconclusive results when the exchangeable phosphorus quantities have been evaluated by chemical methods.

However, this does not mean that these oxides have been inactive in the soils. An assumption is that the quantities involved may have been insufficient. It is very difficult to determine the amount of fixed phosphorus quantity required to decrease the available phosphorus quantity. Indeed, the balances controlling this are extremely complicated. However, our study has clearly demonstrated that the phosphorus quantities present in the soil solution were depressed by the use of iron oxides. It is therefore probable that iron oxides have fixed a good part of phosphorus but that this fixation is not very stable. Phosphorus fixed on these oxides would still be, in this case, still available to the plants or extractable to the chemical extractant.

The chemical extraction method involves ammonium acetate and EDTA and is achieved at pH 4.65. This method can then extract phosphates slightly linked to, for example, iron oxides. The same explanation can be applied to the vegetals which developped mechanisms (protons liberation for example) allowing them absorb other forms of phosphorus than orthophosphates in the soil solution. This is particularly true for vegetals adapted to nutrients-poor soils.

Moreover, it seems well established that the phosphates adsorption on the iron oxides is a two step reaction. A fast stage is followed by a slower second one (Barrow, 1983a). The initial reaction is quick and would be achieved in a few hours (Madrid & De Arambarri 1985) and is a simple ligand exchange ligands. The slow reaction is not well defined and numerous hypothetical models are described (Lookman et al., 1994; Barrow, 1983b; Parfitt, 1989). It seems that this second stage involve the diffusion of phosphate ions inside the iron oxide structure, leading to a more stable relationship between the two elements concerned. It is then possible that, during the experiment, the second reaction between the phosphate ions and the iron oxides was not yet sufficiently advanced and that these phosphate ions were still available to the molecules used for extraction or to the roots of plants adapated to oligotrophic conditions. Moreover the decrease of the soil solution phosphates contents in the samples having received iron oxides seems to confirm this assumption. It is therefore probable that the use, in these conditions, of treated iron oxides (85 times more reactive) would allow better result regarding the soils available phosphorus.

F. CONCLUSION

The creation of an ecological network linking the existing species-rich vegetal herbaceous associations requires the use of green spaces or situated on the road verges. They are often nutrients-rich lands. When these elements, particularly phosphorus, are available in great quantities to plants, species-rich associations cannot survive. The methods commonly used to decrease the available quantities (soil removal, exportation by plants) seem insufficient for practical or kinetic reasons. The addition of iron oxides may constitute a solution to this problem. Iron oxides originating from industrial processes have been tested. These are industrial processes wastes, and are pure and cheap, which is very important. The major advantages regarding their use is that iron oxides are natural phosphorus-fixing compounds in soils.

After having checked the efficiency of the reaction in solution (20 µmoles P fixed / g iron oxide), these oxides have been tested in the fields. The phosphorus quantities present in the soil solution have been strongly decreased by an adjunction of these oxides; this constitutes a very important and encouraging stage. The decrease of available phosphorus quantities by the use of iron oxides could not be demonstrated, either by chemical or biological extraction. However this can probably be explained by the the quantities or reactivity of the iron oxides used. Very simple methods have enable to highly increase the specific area (30 x) of these oxides and consequently their capacity to adsorb phosphated ions (85 x). It is then likely that the use, in such conditions, of these treated iron oxides would yield better regarding the amount of available phosphorus

Applications of these newly synthetised oxides are wider than the decrease of soil available phosphorus. According to the qualities of these productions mentioned above, other uses could be envisaged, i. e. the treatment of phosphated waters.

XI. GERMINATION OF 6 SPECIES CHARACTERISTIC OF SPECIES-RICH GRASSLANDS IN RELATION WITH NUTRIENT STATUS AND WATER POTENTIAL

A. SUMMARY

The germination of six species characteristic of species-rich grasslands has been analysed with different nutrients concentrations or water potential. Demineralized water and complete nutritive solution were considered as references. The concentrations of N, P, K, considered separatly or together, varied in a range similar to that found in the soil solution. Use of Polyethylene glycol 6000 (PEG) to simulate low water potential has also been tested to control the toxic or osmotic effects of the nutrients.

Low water potentials have decreased germination for *Leontodon* sp. and *Leucanthemum vulgare*. This can be seen as an adaptation preventing the seedlings to face dry conditions.

Nitrogen, potassium or phosphorus do not seem to influence directly, in our experiment, the germination behaviour of the tested species.

Nevertheless an indirect influence of nitrogen on the germination of *Leontodon* sp. by reducing it at high concentrations was observed. It is assumed that this influence is indirect through an osmotic effect. Indeed, the same results are obtained with high quantities of N or with quantities of P.E.G simulating the water potential obtained by these N concentrations.

Seedlings coming from small seeds of *Leucanthemum vulgare* and *Pimpinella saxifraga* did no survive with demineralized water. This could be attributed to the low reserve contained in this seeds. This mortality was significantly decreased by a complete nutritive solution, where NPK were removed, especially for *Leucanthemum vulgare*. The deficiency must therefore concern another nutrient than N, P or K.

B. INTRODUCTION

The restoration of species-rich grasslands is of major concern since their dramatic disappearance these last decades. The decrease of the abundance of these communities is now commonly attributed to the intensification of agricultural practices or, at the contrary, to the cessation of management. The restoration must firstly focus on the abiotic conditions that are adequate to the biodiverse communities. This concerns essentially the soil characteristics regarding namely nutrient status and water availability. Afterwards, the biotic conditions have to be considered. It could be assumed that the re-appearance of the new species from the seed bank or the seed rain should be the basis of the restoration. Yet, these presumed sources seem in many case not sufficient. The majority of the considered species do not have a permanent seed bank (Bakker *et al.* 1991; Mc Donald, Bakker & Vegelin 1996; Thompson, Bakker & Bekker 1997). Moreover, in many places, semi-natural grasslands have become so rare that seed rain is not sufficient to ensure the regeneration of new communities.

The restoration has to be considered in many cases by re-introduction of seeds. In a first step, the best conditions of germination and seedling survival have to be studied. It is obvious that the maintenance of species-rich grasslands depends on moderate productivity (Grime 1979; Tilman 1990). The soil nutrient content is known to be inversely related to species diversity. This concerns especially nitrogen (Elisseou *et al.* 1995; Marrs 1993; Tallowin *et al.* 1994; Willems, Peet & Bik 1993) and phosphorus (Janssens *et al.* 1997).

Meanwhile, few studies have concerned the effects of high quantities of these elements on germination and seedlings establishment. This is rather surprising since the restoration of species-rich grasslands often has to be carried out on soil enriched by agricultural practices. Egley & Duke (1985) assume that most inorganic ions do not have any specific effect on germination, nitrate ions being the exceptions. Many studies have indeed shown that nitrogen is a major factor interfering with germination (Karssen & Hilhorst 1993). The promotive action of small concentration of nitrate on germination is known for a long time (Lehman 1909). Yet, the mechanism of this promotive action is still unknown. The ecological significance of the promotion of germination or breaking of dormancy by small quantities of nitrate could be related to an adaptation to promote the germination in the gaps, more favourable to the establishment of seedlings (Pons 1989). Meanwhile, nutrients could inhibit germination non-specifically when they occured in high concentration in the soil (Karssen & Hilhorst 1993). It appears from several studies that high nitrate contents can depress germination (Kitajima & Tilman 1996; Pons T. 1989; Thanos & Rundel 1995).

The initial nutrient concentration in the seeds should have an influence on germination but very little information on the subject is available. It is clear that maternal effects on seeds during development are of major importance (Gutterman 1993), but these effects concerning the nutrients remain unclear. Moreover, seeds of many species loose quantities of soluble substances by leakage (Fenner 1986).

The aim of this paper is to enlighten the influence of nutrients like nitrogen, phosphorus and potassium separatly or together on the germination of 6 species characteristics of species-rich swards. The osmotic effects of these nutrients is also assessed by use of polyethylene glycol 6000 (P.E.G.).

C. MATERIAL AND METHODS

Six species characteristics of the species-rich hay meadows of Belgian Ardennes (Alchemillo-Trisetetum) have been considered :

- *Centaurea gr. Jacea*
- *Leucanthemum vulgare*
- *Leontodon autumnalis*
- *Leontodon hispidus*
- *Pimpinella saxifraga*
- *Plantago lanceolata*

The seeds tested, bought at a seed-trader in Switzerland, were conditionned in dry and dark conditions just after collecting. The experiment has begun 1 year later.

The nutritive solution was made of (table 1) :

Components	ppm
$Ca(NO_3)_2.2H_2O$	47.20
$NaNO_3$	34.00
KCl	18.63
K_2SO_4	21.75
$NaH_2PO_4.2H_2O$	3.12
$MgCl_2.6H_2O$	10.17
$MgSO_4.7H_2O$	12.32
$CaCl_2.2H_2O$	36.75
$CaSO_4.2H_2O$	43.00
Fe EDTA	27.51
H_3BO_3	4.94
$ZnSO_4.7\ H_2O$	0.23
$MnCl_2.4\ H_2O$	1.58
$CuSO_4.5H_2O$	0.20
$(NH_4)_6Mo_7O_{24}.4H_2O$	1.07

Table 1. Component of the nutritive solution.

The concentrations of the macronutrients in mmoles/l in this solution in the complete solution correspond to : N = 0.8; P = 0.02; K = 0.5; Ca = 0.7; Mg = 0.1; Fe = 0.08; S = 0.42; Na = 0.42; Cl = 0.86.

The concentration of the micronutrients in µmoles/l were as follows : B = 80; Zn = 0.8; Mn = 8; Cu = 0.8: Mo = 6.

These ranges of concentrations can be related to soil concentrations commonly found in extensive grasslands soil (Janssens et al., 1996).

This complete nutritive solution was also modified to test the effect of single elements. The treatment consisted in changing the content of N, P and K separately or together in this complete nutritive solution.

A preliminary test was required in a way to be sure that the addition of accompagnying ions did no induce toxic effects.

The water potential of all these solutions has been measured. This water potential was not significantly different between all the solutions (-0.77 bars) except when nitrogen was present in higher quantity than reference (-1.5 bars). In a way to distinguish a direct effect of the nitrogen or an indirect effect of nitrogen (through a lower water potential), another treatment has been carried out with Polyethylene Glycol 6000 added to the initial complete nutritive solution to obtain -1.5 bars.

These treatments will be called as follows (table 2) :

N°	Treatment	symbol
1	Demineralized water	H_2O
2	Nutritive solution	reference
3	Nutritive solution without N	N*0
4	Nutritive solution without P	P*0
5	Nutritive solution without K	K*0
6	Nutritive solution without NPK	(NPK)*0
7	Nutritive solution with N /10	N/10
8	Nutritive solution with P /10	P/10
9	Nutritive solution with K /10	K/10
10	Nutritive solution with NPK /10	(NPK)/10
11	Nutritive solution with N*5	N*5
12	Nutritive solution with P*5	P*5
13	Nutritive solution with K*5	K*5
14	Nutritive solution with NPK*5	(NPK)*5
15	Nutritive solution with N*10	N*10
16	Nutritive solution with P*10	P*10
17	Nutritive solution with K*10	K*10
18	Nutritive solution with NPK*10	(NPK)*10
19	Nutritive solution with P.E.G.	P.E.G.

Table 2. Solutions tested and abbreviations used below in the text.

The seeds were put on small blocks (8.5 cm*6 cm) of expansed polystyrene, floating on the nutritive solution. Each block was covered by filter paper, this filter paper was large enough to soak in the solution in a way to be saturated by it. This experimental scheme allows to always keep the species in contact with the solution without any risk of immersion. Two liters of each solution were used in small containers (20 cm x 22 cm x 10 cm). Six frigolite blocks were put in each container. Thirty seeds were sown on each frigolite block, one species per frigolite block. The six species were thus in contact with the same solution in the same conditions. Water lost through evaporation was compensated with demineralized water. No changes of composition of the diverse solution were observed until the end of experiment by analysis of the water potential, electrical conductivity and pH. Benzimidazole (100 ppm) was use as fungicide, this molecule has a large action spectrum with no influence on germination (Maude 1996).

The experiment was carried out in three replicates in a conditioned room at a constant temperature of 22 °C and a relative humidity of 65%. Light was provided by 12 Metalarc lamps (400 W) with a 12h-daily photoperiod (83.6 W/m^2 at seeds level)

The germinated seeds and the dead seedlings were counted every 2 days during 6 weeks, this period was sufficient to exhaust the seed reserves of the plants. Mortality was calculated as a percentage of dead seedlings versus germinated seeds. T50 was the number of days to obtain 50% of the final percentage of germination. Statistical differences were assessed by one-way ANOVA and Fisher's LSD test.

For each species, 100 seeds were weighed to calculate an average seed weight.

D. RESULTS AND DISCUSSION

All the results are shown in appendice 1

Pimpinella saxifraga and *Leontodon hispidus* are the species that show the highest percentages of germination in demineralized water (table 3). *Centaurea gr. Jacea* has the lower germination percentage. These results are similar with those of Grime (1981) only regarding *Leontodon hispidus, Leontodon autumnalis and Centaurea gr. Jacea;* the results obtained for *Pimpinella saxifraga* are here very different.

species	germination	mortality	T50	Grime '81		weight (mg seed^{-1})
				germin.	T50	
Pimpinella saxifraga	81 ^{20} ab	53 ^{32} a	7 ^{0} abc	15	17	0.61
Leontodon hispidus	77 ^{12} ab	57 ^{13} a	6 $^{1.2}$ abc	74	10	1.19
Leucanthemum vulgare	66 ^{17} abc	66 ^{31} a	6 $^{1.2}$ abc	84	3	0.40
Leontodon autumnalis	63 ^{10} abc	65 ^{21} a	6 $^{1.2}$ abc	50	4	1.25
Plantago lanceolata	51 ^{15} abc	46 ^{40} a	5 ^{0} bc	73	5	1.66
Centaurea gr. Jacea	36 ^{16} bc	42 ^{26} a	8 $^{2.3}$ ab	45	9	2.24

Table 3. Percentage of germination, mortality and T50 of the 6 species with demineralized water. Small figures represent the standard deviation and letters distinguish groups for a Tukey test ($\alpha=0.05$; $n=3$) . The last two columns present the results (percentage of germination and T50) obtained by Grime (1981) and the seeds

weights used in our experiment.

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The germination percentages obtained with the complete nutritive solution are lower than with demineralized water (table 4), but the species are classified in almost the same order (*Plantago lanceolata* replaces *Centaurea gr. Jacea* in the 6th place).

species	germination	mortality	T50
Pimpinella saxifraga	77 ^{15} ab	28 ^{26} abc	11 ^{0} a
Leontodon hispidus	56 $^{5.1}$ abc	70 $^{5.7}$ ab	7 ^{0} a
Leucanthemum vulgare	56 $^{1.9}$ abc	24 ^{15} abc	11 $^{3.5}$ a
Leontodon autumnalis	54 ^{12} abc	73 ^{11} ab	6 $^{1.2}$ a
Plantago lanceolata	34 $^{8.4}$ bcd	37 ^{29} abc	8 $^{2.3}$ a
Centaurea gr. Jacea	16 ^{15} cd	17 ^{29} bc	9 $^{7.2}$ a

Table 4. Germination, mortality and T50 of the 6 species with the reference solution.

a) *Pimpinella saxifraga*

This species exhibits the highest percentages of germination in demineralized water (81%). This was not found by Grime (1981) who obtained 15 % of germination after comparable dry storage and germination conditions; nevertheless, after chilling at 5°C, this germination percentage reached 86%. In that work, species which responded to chilling treatment are comparatively large (except for arable weeds or marsh plants). The seeds of *Pimpinella saxifraga* tested by Grime (1981) where bigger (2.12 mg) than in our sample (0.61 mg). The high germination ability of this species in our study could be explained by the small size of its seeds. Fenner (1983) has found a clear trend for small seeds to have relatively large embryos and thin seed coats and to have seedlings growing faster.

Nutrients do not seem to affect the germination behaviour of this species. The only slight decrease of germination is found when demineralized water is compared with NPK*10. This is also true as far as mortality is concerned. An explanation can be that, at these high concentrations, only seeds with more developped embryos are able to germinate. The subsequent seedlings are then more developped, allowing a higher survival. P.E.G. has also decreased mortality comparatively to N*10. The high mortality of N*10 could be attributed to penetration of N into the seeds (Thanos & Rundel 1995) inducing a toxic effect but this mortality is not different to that obtained with demineralized water or reference solution.

High osmotic potential levels (high N content and P.E.G.) also decreases T50 by probably a delayed imbibition of the seeds.

No any effect of phosphorus or potassium has been enlightened. Concerning phosphorus, this is consistent with Fenner & Lee (1989) who estimated that the amount of this element in seeds is sufficient for the seedlings requirements in pasture grasses and legumes. Fenner & Lee (1989) found nevertheless that seedlings have an immediate requirement for N and K; this was not found in our results. The absence of N and K have indeed not induced an increased mortality.

b) Leontodon autumnalis and Leontodon hispidus

The germination behaviour of *Leontodon hispidus* is very similar to *Leontodon autumnalis.*

Germination is affected by osmotic potential. The treatments N*5, N*10 and P.E.G. show germination percentages significantly lower than demineralized water, reference, N=0 & N/10. P.E.G. has induced the same effect than the N*5 & N*10 treatments allowing to attributed the reduce of germination to osmotic potential.

P.E.G. treatment has decreased mortality comparatively to N*10. Nevertheless, this does not imply a toxic effect of N since differences between all N treatments are not found. The same explanation as for the previous species should thus be applied here. Both treatments (P.E.G. & N*10) increase significantly the T50, which, as for previous species, can be due to a delayed imbibition of the seeds. The same explanation can be applied to decreased T50 by NPK *10 comparatively to treatments without N.

Here again, the germination behaviour of *Leontodon* sp. is not changed by phosphorus or potassium.

c) Leucanthemum vulgare

The germination behaviour of this species is very similar to that of *Pimpinella saxifraga.* The shape and the size (0.40 mg for *Leucanthemum vulgare* and 0.61 mg for *Pimpinella saxifraga)* of the seeds of these species are similar. Because of the small size of its seeds, the germination percentages should be higher than for *Leontodon* sp. It can be argued that the lower germination obtained with our sample is due to the dry storage applied to our seeds (Grime 1981).

The germination potential of this species is only decreased by P.E.G. N*10 has not affected the germination. The negative osmotic effect is thus balanced by the presence of nitrogen. An hypothesis could be that N penetrates seeds without inducing any negative effect. This N and the associated ions in the seeds can thus change the osmotic equilibrium allowing a better imbibition. Another interpretation could relie to dormancy breaking since at low water potential it has been shown that increased light increased dormancy breaking (Pons 1993). In our experiment, light is constant but nitrate is known to be able to replace light in this breaking dormancy (Karssen & Hilhorst, 1993).

Demineralized water has induced a significantly higher mortality comparatively to all other treatments. Though mortality is not decreased, when N, P, K or NPK are absent, comparatively to treatments where they are present. It seems thus that this species needs another element present in the solution to survive. This element could be : Ca, Mg, Fe, S, Na or Cl. The roots of the seedlings growing on demineralized water were pink; that could indicate an iron deficiency (Fenner & Lee 1989).

d) Centaurea gr. Jacea

The germination of this species is lower than the germination of *Pimpinella saxifraga* and *Leontodon hispidus* . The size of the seeds of *Centaurea gr. Jacea* is also bigger (2.24 mg). The treatments do not affect the behaviour of germination.

e) Plantago lanceolata

The percentage of germination of this species is only raised by demineralized water vs all other treatments. This treatment has also largely increased mortality but with a high variability between replicates. It was observed, as in many other studies, that the seeds of this species were covered of mucilage after imbibition. It is possible that the presence of an element in the solution other than NPK has prevented an adequate formation of this mucilage.

E. CONCLUSION

In the conditions of the experiment, nutrients do not seem to influence directly the germination of the species tested. Phosphorus and potassium have not induced any change of germination behaviour by deficiencies or excess. Excess of nitrogen has reduced germination percentage for *Leontodon* sp. This last nutrient is the only one that changes the water potential. When P.E.G. is added to complete nutritive solution (reference) in a way to reach the water potential obtained with high nitrogen concentration, the reduction of germination is similar. The effect of nitrogen must thus be seen through osmotic potential.

Many studies have enlightened the breaking of dormancy by nitrogen (especially nitrate) (Karssen & Hilhorst 1993; Pons 1989; Thanos & Rundel 1995). In our case, nitrogen does never raise germination. This might be due to non dormant seeds or more likely to the light provided, it has indeed been shown that nitrate and light may replace each other in breaking of dormancy (Karssen & Hilhorst, 1993).

Leontodon sp. and *Leucanthemum* vulgare have shown a decrease of germination by use of low water potentials. This has an ecological significance, since rainfall, inducing more favourable conditions, dilutes salts in the soil water. Therefore reduced germination related to lower water potential can be seen as an adaptation to prevent the seedlings to face unfavourable dry conditions (Ungar 1977).

Demineralized water has induced a higher mortality with *Leucanthemum vulgare* and to a lesser extent with *Pimpinella saxifraga.* These species have very small seeds, the reserves are thus very low. The seedlings need very quickly an intput of nutrient to survive. Moreover seeds in demineralized water release their inner soluble nutrient (especially nitrogen) in the surrounding water by leakage (Fenner, 1986). The case of phosphorus is particular since it has been shown that few days after germination mycorrhyza are already established (Gay, Grubb & Hudson 1982). Nevertheless, concerning *Leucanthemum vulgare* the nutritive solution without NPK raises significantly the survival comparatively to demineralized water. The deficiency must therefore concern another nutrient.

Appendice 1. Results obtained for each species.

Centaurea nigra

treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.	
H2O	36	16	a	42	26	a	8	2	bc	H2O	36	16	a	42	26	a	8	2	a	H2O	36	16	a	42	26	a	8	2	a	H2O	36	16	abc	42	26	a	8	2	a
N=0	33	7	a	27	3	a	9	7	abc	P=0	41	10	a	28	16	a	6	1	a	K=0	41	13	a	34	19	a	9	3	a	NPK=0	41	2	ab	13	12	a	7	0	a
N/10	37	3	a	36	17	a	6	1	bc	P/10	29	13	a	43	29	a	8	3	a	K/10	38	5	a	32	12	a	10	2	a	NPK/10	30	6	abc	47	26	a	9	5	a
Ref.	34	8	a	37	29	a	8	2	bc	Ref.	34	8	a	37	29	a	8	2	a	Ref.	34	8	a	37	29	a	8	2	a	Ref.	34	8	abc	37	29	a	8	2	a
N*5	38	7	a	41	26	a	11	4	abc	P*5	34	10	a	33	21	a	7	0	a	K*5	36	5	a	25	3	a	6	1	a	NPK*5	23	3	abc	25	11	a	11	6	a
N*10	27	19	a	18	17	a	14	6	abc	P*10	37	3	a	19	20	a	7	0	a	K*10	28	8	a	24	11	a	9	5	a	NPK*10	19	2	bc	12	11	a	10	8	a
P.E.G.	22	10	a	13	12	a	18	2	ab																														

Leontodon autumnalis

treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.	
H2O	63	10	ab	65	21	abc	6	1	b	H2O	63	10	a	65	21	a	6	1	a	H2O	63	10	a	65	21	a	6	1	a	H2O	63	10	a	65	21	a	6	1	bc
N=0	69	11	ab	67	14	abc	7	0	b	P=0	44	13	a	65	5	a	7	0	a	K=0	62	4	a	55	24	a	6	1	a	NPK=0	68	11	a	67	9	a	6	1	bc
N/10	69	13	ab	67	9	abc	8	3	b	P/10	52	16	a	65	40	a	7	0	a	K/10	58	14	a	62	25	a	10	6	a	NPK/10	64	5	a	54	15	a	7	0	abc
Ref.	56	5	abc	70	6	abc	7	0	b	Ref.	56	5	a	70	6	a	7	0	a	Ref.	56	5	a	70	6	a	7	0	a	Ref.	56	5	a	70	6	a	7	0	abc
N*5	30	17	cd	64	4	abc	8	3	b	P*5	44	14	a	48	19	a	7	0	a	K*5	49	7	a	68	17	a	6	1	a	NPK*5	48	11	a	67	15	a	8	2	abc
N*10	32	2	bcd	80	20	ab	14	6	a	P*10	56	8	a	69	10	a	7	3	a	K*10	62	7	a	51	19	a	6	1	a	NPK*10	47	12	a	54	19	a	13	5	ab
P.E.G.	39	16	bcd	34	9	bc	15	3	a																														

Leontodon hispidus

treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.	
H2O	77	12	ab	57	13	ab	6	1	bcd	H2O	77	12	ab	57	13	a	6	1	a	H2O	77	12	ab	57	13	a	6	1	a	H2O	77	12	ab	57	13	a	6	1	a
N=0	66	23	ab	61	17	ab	6	1	cd	P=0	51	4	bcd	55	10	a	6	1	a	K=0	64	7	abc	64	2	a	6	1	a	NPK=0	66	7	abc	62	10	a	5	0	a
N/10	67	7	ab	54	13	abc	6	1	cd	P/10	50	12	bcd	53	16	a	10	6	a	K/10	72	8	abc	54	20	a	7	0	a	NPK/10	57	12	abc	64	7	a	7	0	a
Ref.	54	12	abcd	73	11	ab	6	1	cd	Ref.	54	12	abcd	73	11	a	6	1	a	Ref.	54	12	abc	73	11	a	6	1	a	Ref.	54	12	abc	73	11	a	6	1	a
N*5	36	10	bcde	67	12	ab	13	7	abc	P*5	54	14	abcd	54	13	a	6	1	a	K*5	49	4	bc	62	15	a	7	3	a	NPK*5	52	8	bc	54	14	a	10	2	a
N*10	27	7	cde	71	4	ab	7	3	bcd	P*10	46	10	bcd	45	12	a	7	0	a	K*10	51	12	bc	60	25	a	6	1	a	NPK*10	44	5	bc	65	14	a	11	4	a
P.E.G.	39	8	bcde	25	16	bc	19	4	ab																														

Leucanthemum vulgare

treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.	
H2O	66	17	a	66	31	a	6	1	def	H2O	66	17	a	66	31	a	6	1	a	H2O	66	17	a	66	31	a	6	1	a	H2O	66	17	abc	66	31	a	6	1	bc
N=0	60	15	a	18	6	b	7	0	cdef	P=0	54	5	a	39	7	b	8	2	a	K=0	67	0	a	17	13	b	11	4	a	NPK=0	73	6	ab	14	9	b	8	2	abc
N/10	59	8	a	28	9	b	11	0	bcdef	P/10	51	13	a	7	13	b	13	7	a	K/10	61	7	a	28	9	b	10	2	a	NPK/10	62	5	abc	29	8	b	8	2	abc
Ref.	56	2	a	24	15	b	11	4	abcdef	Ref.	56	2	a	24	15	b	11	4	a	Ref.	56	2	a	24	15	b	11	4	a	Ref.	56	2	abc	24	15	b	11	4	abc
N*5	52	5	a	17	10	b	15	3	abcde	P*5	59	8	a	17	4	b	10	2	a	K*5	44	7	a	14	5	b	13	3	a	NPK*5	50	3	abc	17	15	b	15	3	ab
N*10	53	6	a	21	5	b	17	0	abcd	P*10	64	5	a	23	17	b	8	2	a	K*10	61	5	a	8	14	b	12	2	a	NPK*10	46	26	bc	21	17	b	16	9	ab
P.E.G.	21	4	b	11	10	b	20	1	abc																														

Pimpinella saxifraga

treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.	
H2O	81	20	a	53	32	ab	7	0	cde	H2O	81	20	a	53	32	ab	7	0	a	H2O	81	20	a	53	32	a	7	0	a	H2O	81	20	ab	53	32	ab	7	0	bcd
N=0	82	4	a	30	3	abc	8	2	cde	P=0	72	12	a	37	9	abc	12	5	a	K=0	73	9	a	28	9	a	11	4	a	NPK=0	78	10	abc	41	17	abc	7	0	cd
N/10	73	9	a	36	2	abc	8	2	cde	P/10	71	19	a	42	8	abc	13	3	a	K/10	66	13	a	33	13	a	12	5	a	NPK/10	76	5	abc	27	6	abc	8	2	bcd
Ref.	77	15	a	28	26	abc	11	0	bcde	Ref.	77	15	a	28	26	abc	11	0	a	Ref.	77	15	a	28	26	a	11	0	a	Ref.	77	15	abc	28	26	abc	11	0	bcd
N*5	50	12	a	31	14	abc	16	5	abcd	P*5	79	17	a	18	26	bc	10	2	a	K*5	66	11	a	38	11	a	9	3	a	NPK*5	60	3	abc	22	4	abc	14	3	abc
N*10	57	12	a	58	25	ab	14	3	abcde	P*10	67	6	a	22	11	abc	8	2	a	K*10	74	13	a	34	4	a	12	2	a	NPK*10	49	15	bc	17	18	bc	18	2	ab
P.E.G.	60	19	a	4	8	bc	21	4	abc																														

Plantago lanceolata

treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.		treatment	% germ.	s.d.		% dead	s.d.		T50	s.d.	
H2O	51	15	a	46	40	ab	5	0	bc	H2O	51	15	a	46	40	ab	5	0	bc	H2O	51	15	a	46	40	ab	5	0	a	H2O	51	15	a	46	40	ab	5	0	a
N=0	18	10	b	0	0	bc	8	5	abc	P=0	27	9	b	0	0	bc	13	7	abc	K=0	16	7	b	10	16	bc	10	6	a	NPK=0	22	13	b	18	31	abc	15	16	a
N/10	17	6	b	0	0	bc	17	3	abc	P/10	18	2	b	7	12	bc	21	5	ab	K/10	8	2	b	0	0	bc	6	1	a	NPK/10	21	8	b	0	0	bc	13	10	a
Ref.	16	15	b	17	29	abc	9	7	abc	Ref.	16	15	b	17	29	abc	9	7	abc	Ref.	16	15	b	17	29	abc	9	7	a	Ref.	16	15	b	17	29	abc	9	7	a
N*5	19	4	b	5	8	bc	21	3	ab	P*5	22	7	b	0	0	bc	13	7	abc	K*5	8	8	b	7	12	bc	9	3	a	NPK*5	21	2	b	6	10	bc	15	4	a
N*10	22	8	b	4	7	bc	15	10	abc	P*10	14	4	b	7	12	bc	18	1	abc	K*10	8	5	b	8	14	bc	11	11	a	NPK*10	22	8	b	26	16	abc	20	5	a
P.E.G.	8	8	b	0	0	bc	24	0	ab																														

Legend : % germ. : percentage of germinated seeds
% dead : percentage of dead seeds
T50 : number of days to obtain 50 % of the final percentage of germination

The figures present the means and the standard deviation (s.d.)
The letters distinguish groups for a Tukey test ($\alpha=0.05$; n=3)