ACCOUNTS
of chemical research

Chemically Engineered Extracts: Source of Bioactive Compounds

I. AYELEN RAMALLO, MARIO O. SALAZAR, LUCIANA MENDEZ, AND RICARDO L. E. FURLAN*

Facultad de Ciencias Bioquímicas y Farmacéuticas, Universidad Nacional de Rosario and Instituto de Química de Rosario (Conicet-UNR), Suipacha 531, 2000 Rosario, Argentina

RECEIVED ON JULY 28, 2010

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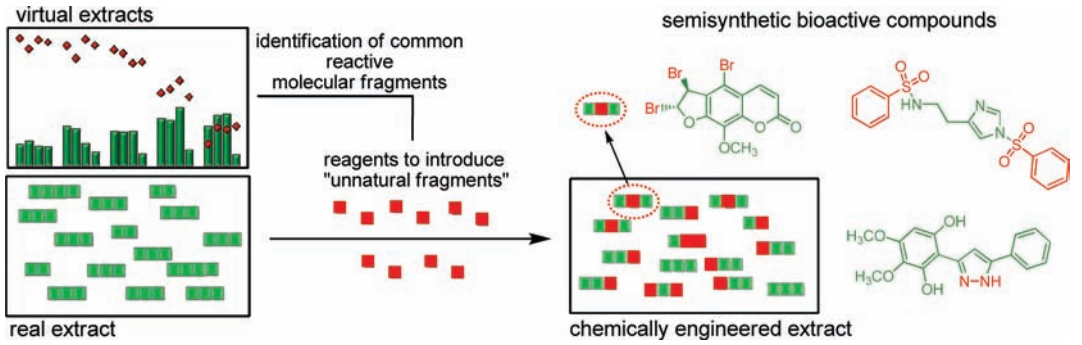

Biological research and drug discovery critically depend on access to libraries of small molecules that have an affinity for biomacromolecules. By virtue of their sustained success as sources of lead compounds, natural products are recognized as "privileged" starting points in structural space for library development. Compared with synthetic compounds, natural products have distinguishing structural properties; indeed, researchers have begun to quantify and catalog the differences between the two classes of molecules. Measurable differences in the number of chiral centers, the degree of saturation, the presence of aromatic rings, and the number of the various heteroatoms are among the chief distinctions between natural and synthetic compounds. Natural products also include a significant proportion of recurring molecular scaffolds that are not present in currently marketed drugs: the bioactivity of these natural substructures has been refined over the long process of evolution.

In this Account, we present our research aimed at preparing libraries of semisynthetic compounds, or chemically engineered extracts (CEEs), through chemical diversification of natural products mixtures. The approach relies on the power of numbers, that is, in the chemical alteration of a sizable fraction of the starting complex mixture. Major changes in composition can be achieved through the chemical transformation of reactive molecular fragments that are found in most natural products. If such fragments are common enough, their transformation represents an entry point for chemically altering a high proportion of the components of crude natural extracts.

We have searched for common reactive fragments in the Dictionary of Natural Products (CRC Press) and identified several functional groups that are expected to be present in a large fraction of the components of an average natural crude extract. To date, we have used reactions that incorporate (i) nitrogen atoms through carbonyl groups, (ii) sulfur by transformation of −OH and amines, and (iii) bromine through double bonds and aromatic rings. The resulting CEEs had different composition and biomolecular properties than their natural progenitors. We isolated a semisynthetic β-glucosidase inhibitor from a CEE prepared by reaction with benzenesulfonyl chloride, an antifungal pyrazole from a CEE prepared by reaction with hydrazine, and an acetylcholinesterase inhibitor from a CEE prepared through bromination. Our results illustrate how biological activity can be generated through chemical diversification of natural product mixtures. Moreover, the level of control that can be asserted in the process by judicious design and experimental choices underscores the potential for further development of CEEs in both basic research and drug discovery.

Introduction

The access to libraries of molecules with interesting biomolecular properties is a limiting step in the drug discovery process. Natural products scaffolds have been invaluable as biologically validated platforms for developing drugs.[1−7] In a series of review articles, Newman, Cragg, and colleagues[2,3,7]

Published on the Web 02/28/2011 www.pubs.acs.org/accounts
10.1021/ar100106n

have analyzed the sources of drugs in the last 30 years. The analysis demonstrated the continuing and valuable contributions of Nature as a source of lead compounds that have provided the basis and inspiration for the synthesis of new drugs.[5,7]

Given this sustained success of natural products (NPs) as sources of bioactive compounds, several research groups have analyzed the differences between natural products, synthetic compounds, combinatorial compounds, and bioactive compounds using cheminformatics tools. In a pioneering work, Henkel et al. found clear differences in molecular properties and structural features between NPs and synthetic molecules.[8] Stahura et al. recognized a set of descriptors that were able to distinguish natural products from synthetic compounds based on their Shannon entropy.[9] Lee and Schneider identified several natural scaffold architectures that were not present in marketed drugs.[10] Feher and Schmidt compared the distribution of a variety of molecular properties among NPs, drugs, and combinatorial molecules, finding that the number of chiral centers, the presence of aromatic rings, the degree of saturation, and the number of various heteroatoms are the most important differences.[11] Ertl and Schuffenhauer performed a cheminformatics analysis of a large collection of natural product structures and compared physicochemical properties and their typical structural features with those of bioactive molecules and average organic molecules.[12] Natural product molecules are more compact (less flexible) because skeletons of natural products are often formed by complex fused rigid ring systems. Natural products contain more oxygen atoms and less nitrogen atoms than the compounds from the two other sets. A lower number of aromatic rings in each structure is also one of the most distinct features of natural products and separates them from other classes of molecules.

In addition to their distinctive diversity, NPs contain numerous bioactive substructures validated by nature's long evolution.[13] A biology oriented synthesis based on this fact has been introduced by Waldmann et al.,[14−18] which builds on a diversity created by nature and aims at its local extension in areas of proven biological relevance using as a starting point simplified core structures of NPs.

Quinn et al. studied the correlation between biosynthetic enzyme/target and observed that an imprint of recognition of protein surfaces during biosynthesis is transferred to recognition of therapeutically useful enzyme targets.[19,20] This biosynthetic enzyme/target correlation provides the underlying reason why natural products are validated starting points for drug design and explains the success of compound libraries based on natural product substructures.[16−18]

Consequently, natural products have become a source of inspiration for the design of natural product-like combinatorial libraries.[21] In parallel, several approaches have been proposed to increase the diversity of natural product mixtures such us their diversification by combinatorial biosynthesis[22−24] and related techniques.[25−28]

Chemical Alteration of Plant Extracts

Chemical alteration of the components of crude natural extracts is an evident way for altering their properties. In fact, there is a series of examples in the literature wherein unintentional formation of solvent artifacts leads to changes in the bioactivity of natural products in a negative or positive way.[29]

In the field of food chemistry, several reports describe the positive effect of hydrolysis on the antioxidant properties of different plant extracts. The phytochemical and antioxidant characteristics of aqueous-methanol extracts from commonly used culinary herbs such as rosemary (*Rosmarinus officinalis* L.), savory (*Satureja hortensis* L.), and thyme (*Thymus vulgaris* L.) have been altered by acid treatment.[30] In general, the process improved the antioxidant potency of the extracts (Figure 1). The authors proposed that some glycosides were transformed to their corresponding aglycones increasing the proportion of phenolic components. As a consequence, the hydrolyzed extracts are more potent participants in redox reactions. This type of process may have an impact upon improving the antioxidative quality of ingredients used for the functionalization of foods and beverages or the manufacture of nutraceuticals and thus further their health-promoting activity.

In a similar study, the effect of acid treatment on the antioxidant properties of extracts of the Taiwanese medicinal plant *Anoectochilus formosanus* was evaluated.[31] *A. formosanus* Hayata (Orchidaceae) has been used popularly as a nutraceutical herbal tea in Taiwan and other Asian countries. Metabolite profiling showed that the aglycones **1**−**3** (Figure 2) of flavonoid glycosides were produced after acid hydrolytic treatment, and this resulted in a significant increase in antioxidant properties of acid-hydrolyzed BuOH fractions.

Acid hydrolysis also improved the antifungal activity of a saponin-rich extract of *Phytolacca dioica* L. (Phytolaccaceae) berries. The activity of the extract was negligible, but the hydrolysate, containing the sapogenins, showed promising antifungal potency against clinical isolates of *Candida albicans* and *Cryptococcus neoformans*.[32]

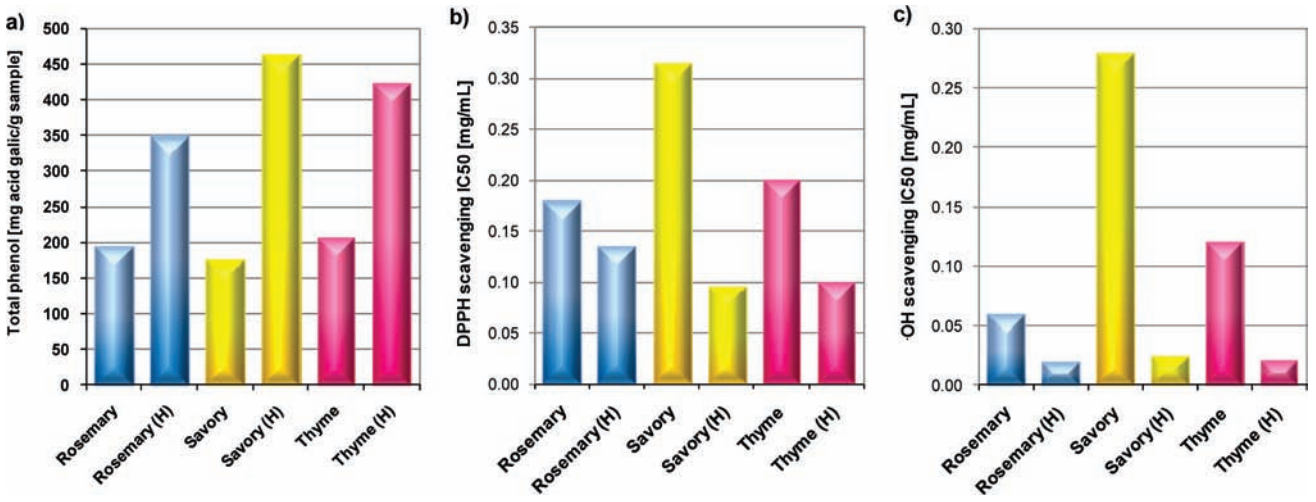

FIGURE 1. The (a) total phenol content, (b) DPPH-scavenging, and (c) hydroxyl radical-scavenging performance for the crude and acid-treated (H) extracts.

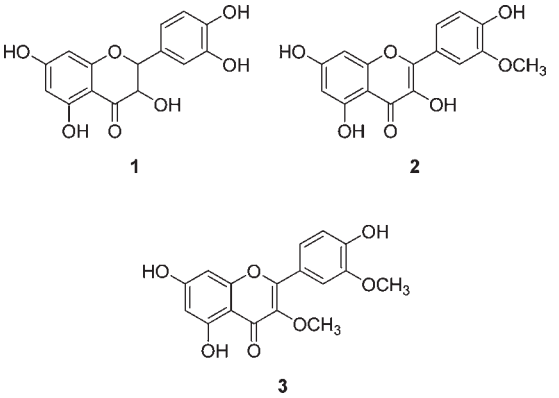

FIGURE 2. Structures of aglycones from *A. formosanus*: dihydroquercetin (**1**); isorhamnetin (**2**); quercetin 3,3′-dimethyl ether (**3**).

Chemically Engineered Extracts

The preparation of chemically engineered extracts (CEEs) through directed chemical diversification of natural extracts (NEs) represents an alternative way to use known and unknown natural scaffolds as starting material for the preparation of semisynthetic compounds libraries. The approach relies on the chemical alteration of a significant proportion of the components of the starting extract: the bigger the number of transformed molecules in the mixture, the higher the chances of generating a compound with interesting biomolecular properties (Figure 3).

Since natural product extracts are mostly uncharacterized libraries of complex composition[33] that usually contain a high number of molecules with different scaffolds and functionalities, the chemical transformation of a high proportion of their component molecules can be challenging. Key for success is the selection of reactive molecular fragments that can be found in many natural small molecules. If such fragments are common enough to be present in a substantial proportion of natural products, their chemical transformation will represent an entry point toward the chemical alteration of a high proportion of crude natural extracts components.

We have searched for such reactive fragments in the CRC Dictionary of Natural Products (DNP version 2001) and observed that various functional groups were present in a high number of the molecules present in the database. According to the DNP, around 75% of the structures within the database contain at least one −OH group, and a similar proportion of molecules include a carbonyl group (73%) or a double bond (65%) (Figure 4). To a lesser extent, natural products also include aromatic rings and nitrogen-containing fragments in their structure (40% and 23%, respectively).

To gain insight into the distribution of those structures within the database, the number of molecules containing these functional groups was estimated in four sets of 17 groups of compounds or "virtual extracts" selected from the database through the use of four different filters. In the first set (S1), each group of compounds contained every molecule within the database found in a particular plant species. Since the number of compounds described for each plant species is rather low, we complemented the data with a second set (S2) where each group of compounds contained every molecule within the database found in one particular plant genus. In the third set (S3), each "virtual extract" included only one type of secondary metabolite (i.e., flavonoids, alkaloids, etc.). This filter was used in order to analyze the distribution of the analyzed functional groups within the different types of metabolites. In set four (S4), each virtual

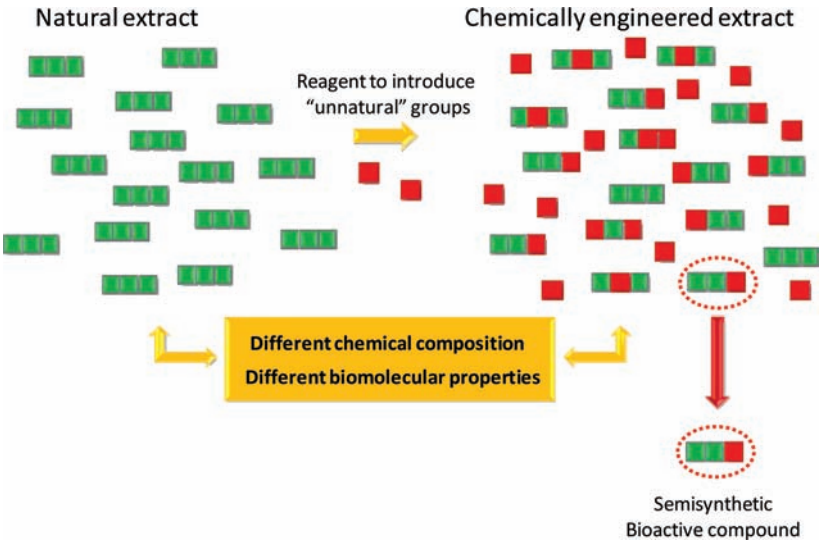

FIGURE 3. Generation of chemically engineered extracts.

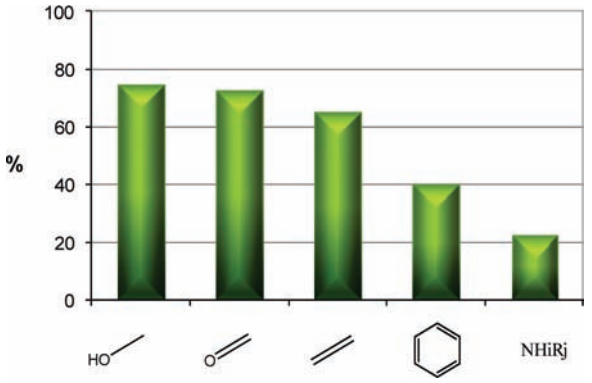

FIGURE 4. Percentages of molecules within the DNP that contain the fragments shown.

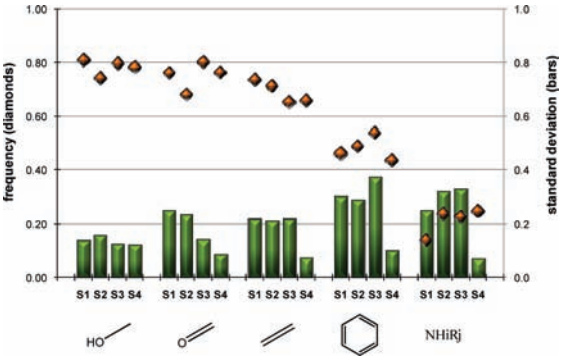

FIGURE 5. Standard deviations (bars) and frequencies (diamonds) of structures containing the major functional groups found in the natural products (C═O, −OH, C═C, aromatic ring, and $-NH_iR_j$ ($i + j = 3$) in the four sets of "virtual extracts": plant species (S1), plant genus (S2), type of metabolite (S3), and molecular weight (S4).

extract included all the molecules in the database with molecular weights starting with arbitrarily defined values (i.e., 10, 20, etc.). This filter generated virtual extracts by grouping molecules using a characteristic that is unrelated to the functional groups that they contain.

As can be observed in Figure 5, the average frequency of molecules containing carbonyls, −OH and double bonds was between 0.65 and 0.81 for all of the sets of virtual extracts. According to these numbers, one could expect that more than half of the different components of a given extract will contain these groups. In addition, there is >80% probability that at least 63% of structures included in the virtual extracts contain one or more carbonyl groups, that at least 69% of the structures contain one or more −OH, and that 57% or more structures include at least one double bond regardless of the filter used. This indicates that the proportion of components that contain these groups does not change in a significant way from one virtual extract to the others. Accordingly, the chemical transformation of any of these three groups could be an interesting entry point to the chemical transformation of a significant number of the component molecules of natural extracts.

A different situation was observed for nitrogen compounds. The average frequency of molecules containing at least once the group $-NH_iR_j$ ($i + j = 3$) was between 0.14 and 0.25, and the standard deviation observed for nitrogen-containing groups was relatively low or high depending on the filter used. The observed frequencies for aromatic rings were higher than those for amines and lower than those for the rest of the functional groups analyzed. Thus chemical transformation of aromatic rings or amines may not be attractive to alter the composition of natural extracts in a significant way. However, the use of reactions that transform

some of the high-frequency functional groups and at the same time transform some of these chemical characters could be interesting.

Transformation of common chemical functionalities gives access to alteration of a significant proportion of natural extract components. In addition, if those functionalities were transformed into chemical groups that are rarely produced by nature, we could complement nature's synthetic capabilities. To increase the chances of generating libraries with interesting biomolecular properties, the approach also attempts to incorporate some characteristics of biologically active compounds into the natural mixtures (i.e., average heteroatom content).

To date, we have used reactions that incorporate nitrogen atoms through carbonyl groups, sulfur by transformation of −OH and amines, and bromine by reaction with double bonds and aromatic rings.

Aiming at the diversification of the components of natural extracts through the chemical transformation of −OH and amines, we have tested the reaction with arylsulfonyl chlorides. The reaction results in the introduction of one (or more) relatively big group into the molecule through the exchange of one hydrogen atom for an arylsulfonyl moiety. This moiety acts as a spectroscopic tag to analyze the alteration in the chemical composition introduced through the reaction. This is particularly important if we take into account that both the starting natural extract composition and the CEE composition are unknown.

When a group of 11 crude plant extracts were treated with *p*-toluenesulfonyl chloride, significant differences in composition were observed. GC−MS and LC−UV chromatograms of the CEEs were qualitatively and quantitatively different from chromatograms of the natural extracts: most of the peaks observed in the chromatograms of each NE disappeared after the reaction and most of the peaks present in the chromatograms of each CEE are absent in the corresponding NE chromatogram (Figure 6).[34]

Changes in composition of the mixtures were also evident from ^{1}H NMR coupled to principal component analysis (PCA). The score plot showed discrimination between the two groups by principal components (PCs), 1 and 2 (Figure 7). CEEs showed a positive PC2 value mainly due to the positive effect on PC2 of the signals corresponding to the *p*-toluene moiety introduced to the natural components of the starting mixtures.

Furthermore, the areas for those signals in the spectra of CEEs and NEs showed significant differences for all the samples: on average, the relative areas of the signals are six times bigger in the CEEs than in the corresponding natural extracts (Figure 8).

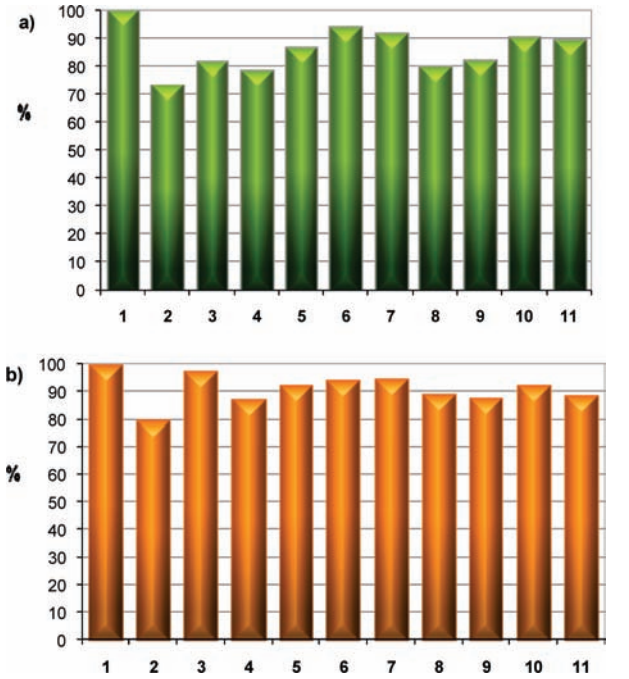

FIGURE 6. Comparison of the chromatograms of 11 NEs and 11 CEEs: (a) percentage of peaks present only in the NEs; (b) percentage of peaks present only in the CEEs; (1) *Conium maculatum* L. (Apiaceae), (2) *Urtica urens* L. (Urticaceae), (3) *Solanum diflorum* Vell. (Solanaceae), (4) *Morrenia brachystephana* Griseb. (Asclepiadaceae), (5) *Matricaria recutita* L. (Asteraceae), (6) *Sonchus oleraceus* L. (Asteraceae), (7) *Brassica rapa* L. (Brassicaceae), (8) *Solanum sisymbriifolium* Lam. *var. sisymbriifolium* (Solanaceae), (9) *Commelina erecta* L. var. erecta (Commelinaceae), (10) *Oenothera affinis* Cambess. (Onagraceae), and (11) *Sida rhombifolia* L. (Malvaceae).

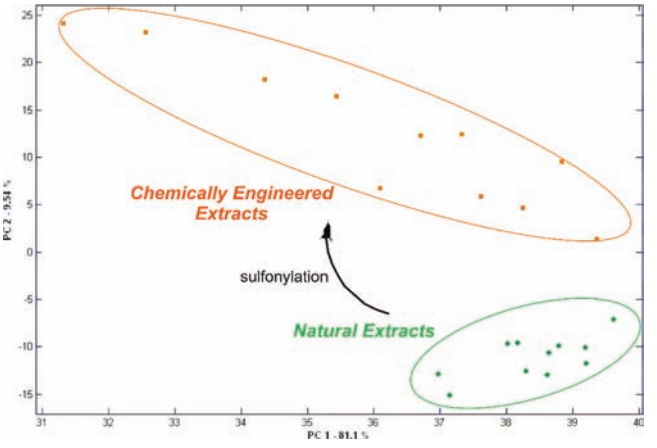

FIGURE 7. Score plot of PCA of ^{1}H NMR data from 11 natural extracts and 11 chemically engineered extracts produced by reaction with *p*-toluenesulfonyl chloride. Natural extracts (♦) and chemically engineered extracts (■).

Considering the differences in composition observed between the natural extracts and the CEEs, it can be expected that

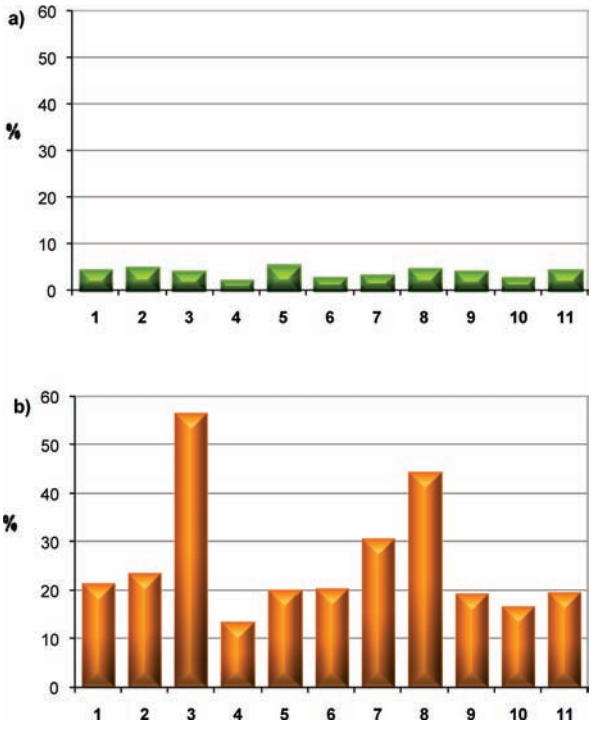

FIGURE 8. Percentage of the total integration of the ^{1}H NMR spectra of extracts corresponding to the chemical shift range 2.20−2.50 ppm and 7.20−7.80 ppm: (a) natural extracts; (b) chemically engineered extracts.

the interaction of these mixtures with biomolecules will also be affected by the reactions. Biomolecular properties of sulfonylated and natural extracts were compared by TLC bioautography a technique particularly suited for the analysis of mixtures. This methodology allows the evaluation of inhibitory properties of a sample spotted onto a TLC plate covered with a gel that contains enzyme, substrate, and a revealing reagent for the product. Using this methodology we investigated inhibitory activities of the enzymes acetylcholinesterase,[35] xanthine oxidase,[36] and β-glucosidase,[37] all involved in different pathological processes. The chemically engineered extract produced by reaction of the plant *Urtica urens* L. (Utricaceae) with benzenesulfonyl chloride showed some interesting differences in its biomolecular properties compared with the starting natural extract. β-Glucosidase TLC bioautography showed a new inhibition spot present in the CEE and absent in the starting NE. The agent responsible for the bioactivity was the dibenzenesulfonyl histamine derivative **4** (Figure 9).[38] This compound includes one natural histamine moiety and two benzenesulfonyl moieties introduced during the diversification step. Although the phytochemistry of *U. urens* is still largely unexplored, the presence of histamine has been reported in the glandular hairs of several species of nettle.[39] To confirm the origin of **4**, histamine was treated with benzenesulfonyl

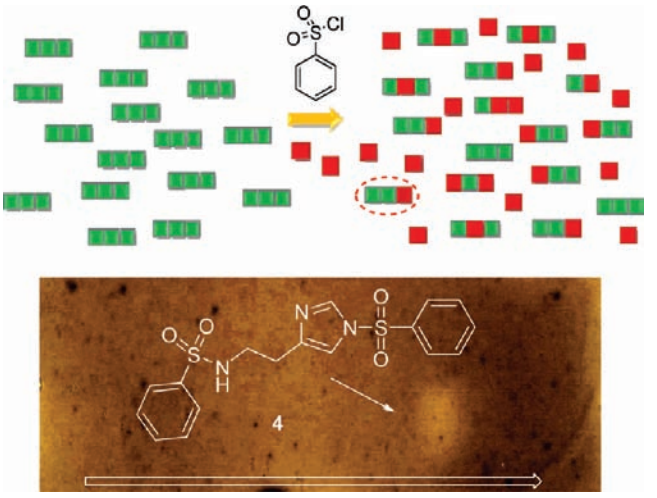

FIGURE 9. β-Glucosidase inhibition by semisynthetic compound **4** isolated from the CEE prepared by reaction of the NE of the plant *Urtica urens* with benzenesulfonyl chloride.

chloride in the same conditions previously used for the sulfonylation of the plant extract, and **4** was the main product obtained. The natural starting material histamine is at least 100 times less active than the generated semisynthetic compound.

In order to alter the composition of natural extracts through chemical transformation of carbonyl groups, we tested the reaction with hydrazine monohydrate. This reagent can react with different carbonyl-containing groups to form hydrazones or acyl hydrazides producing the exchange of one oxygen atom with two nitrogen atoms. Such exchange is interesting considering that different reports indicate that the average nitrogen content per molecule in natural products is lower than that in drug molecules, and the opposite is observed for oxygen content.[8,10,11] The second nitrogen atom in the reagent is attractive because it can give further reactions increasing the potential number of products, and the N−N moiety is uncommon in natural products (only 3% of the natural products that contain nitrogen included in the DNP contain two nitrogen atoms attached to each other).

When an extract of the plant *Polygonum ferrugineum* Wedd. (Polygonaceae)[40] was treated with hydrazine monohydrate, interesting changes were observed by ^{13}C NMR analysis (Figure 10). The main signals that disappear from the spectrum of the NE because of the reaction were located in a region where carbonyl carbon signals of ketones, esters, aldehydes, and amides are expected (between 170 and 210 ppm). In addition, several signals emerge between 150 and 175 ppm, the range of chemical shift of the ^{13}C NMR where

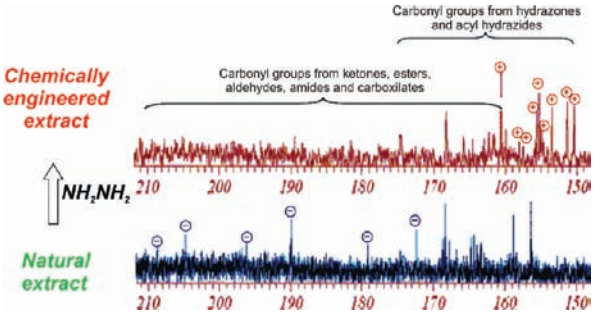

FIGURE 10. Carbon carbonyl region of the ^{13}C NMR spectra of BuOH extract of *P. ferrugineum* after reaction with hydrazine monohydrate and BuOH extract of *P. ferrugineum*..

C=N carbon signals of hydrazones and C=O carbon signals of acyl hydrazides are expected to appear.

As predicted by the calculation with virtual extracts, transformation of the carbonyl groups present in different molecules of the extract introduced significant changes in the composition that could be observed by HPLC and TLC (Figure 11a). The reaction with hydrazine also introduced changes in the antifungal properties of the extract. Bioautography with *Candida albicans*,[41] the most frequently isolated human fungal pathogen,[42,43] showed a clear inhibition zone detected only in the CEE (Figure 11b). Bioactivity-guided fractionation of the semisynthetic mixture led to the isolation of pyrazole **5** that contains the expected N−N moiety in its structure. Pyrazoles are very uncommon secondary metabolites in plants,[44] so pyrazole **5** could be formed by reaction of hydrazine with an inactive flavone present in the unmodified extract.[45,46]

Aiming at the diversification of the components of natural extracts through chemical alteration of aromatic rings and double bonds, we tested a bromination reaction. Most of the natural organobromine compounds are produced by marine organisms,[47,48] and several brominated metabolites with antibacterial,[49] antitumor,[50] antiviral,[51] and antifungal activity[52] have been isolated from seaweeds, sponges, corals, molluscs and others. In contrast, terrestrial plants account only for a few bromine-containing compounds.[47,48] Therefore, we thought about modifying natural scaffolds present in herbal extracts from terrestrial plants through bromination and then analyzing the effect on the biomolecular properties of the mixtures. Considering the target molecular fragments, double bonds and aromatic rings, addition and substitution reactions could be expected. The process would result in the exchange of one hydrogen atom with one bromine atom or in the addition of a bromine atom to a sp^2 carbon atom. Both changes increase the hydrophobicity and

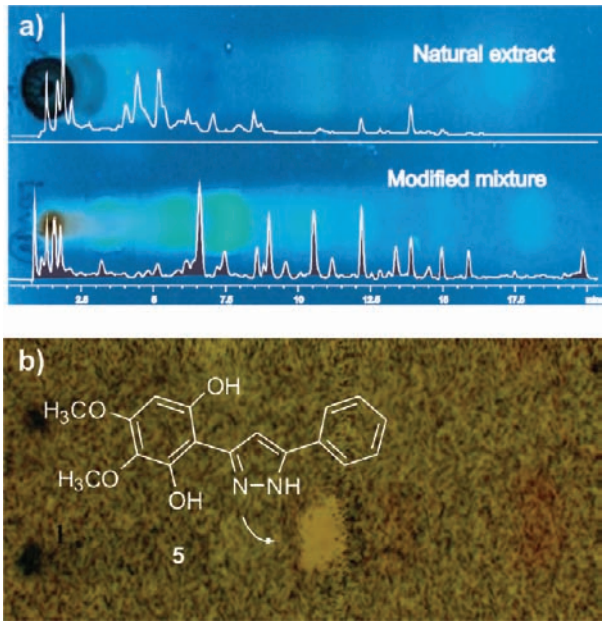

FIGURE 11. (a) Changes in chromatographic profiles produced by reaction of *P. ferrugineum* NE with hydrazine. (b) Antifungal activity of semisynthetic compound **5** isolated from *P. ferrugineum* CEE.

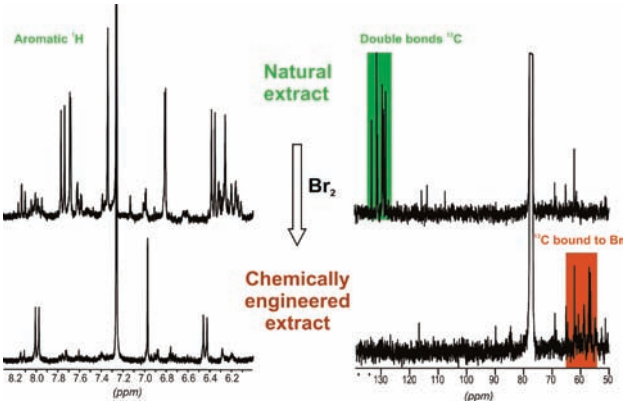

FIGURE 12. Aromatic region of ^1H NMR spectra and part of the ^{13}C NMR spectra of *C. maculatum* natural extract and of *C. maculatum* extract after reaction with bromine.

the size of the molecule, affecting as well its shape and electronic properties. Incorporation of halogen atoms in drug leads is a common strategy to modify molecules in order to vary their bioactivities and specificities,[53] moreover several studies demonstrate that the average proportion of bromine in drugs is significantly higher than that in natural products.[8,11,54]

When the methanol crude extract of the weed *Conium maculatum* L. was treated with bromine, interesting changes were observed in composition and in biomolecular properties.[55] The transformation of both target groups, double bonds and aromatic rings, was suggested by NMR analysis (Figure 12). Double bond characteristic signals

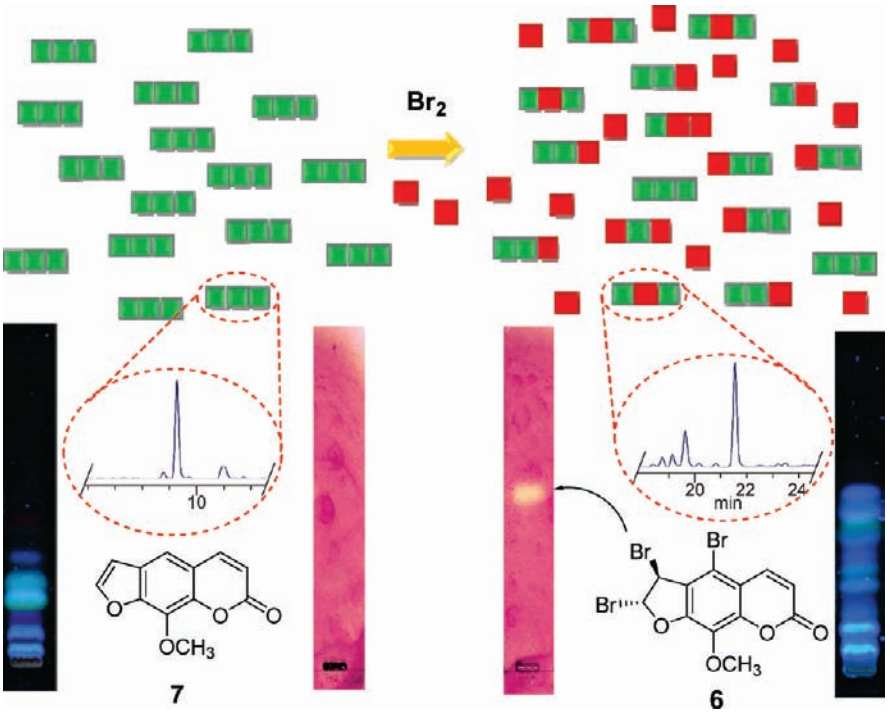

FIGURE 13. Acetylcholinesterase inhibition by semisynthetic compound **6** isolated from the CEE prepared by reaction of *C. maculatum* NE with bromine. Compound **6** is produced from compound **7** present in the NE.

disappear from the ^{13}C NMR spectrum after the reaction, whereas signals in the expected region for sp^3 carbons bound to bromine appear. In addition, the reaction produces a clear decrease of the aromatic signals in the ^1H NMR spectrum.

Bromination of the NE produced a CEE that inhibited the enzyme acetylcholinesterase, a therapeutic target for Alzheimer's disease.[56] When a developed TLC of the NE and CEE was stained by an enzymatically generated dye,[35] a clear inhibition zone was detected in the modified extract, whereas no inhibition was detected in the natural extract (Figure 13). Bioactivity-guided chromatography of the semisynthetic mixture led to the isolation of compound **6**, which contains three bromine atoms in its structure. This brominated derivative is formed from the natural component xanthotoxin (**7**), one of the many components of the natural extract. The peak corresponding to compound **7** disappears from the HPLC-UV chromatogram of the natural extract due to the reaction, and a new peak appears that corresponds to the brominated compound **6**.

Interestingly, the structure of **7** includes the two target functionalities (aromatic ring and double bounds) and both reacted with bromine to produce **6**. The inhibitory potency of **6** and **7** was compared with that of known acetylcholinesterase inhibitors using TLC and microplate assays. Compound **7** was inactive in the range of concentration tested with a pMIQ value below 9.4. Quite the opposite, compound **6** showed a pMIQ of 10.65, which is within the activities observed for some of the best known inhibitors for the enzyme such us physostigmine (pMIQ = 11.81) and galanthamine (pMIQ = 10.57).[57] The pIC_{50} observed for compound **1** in the microplate assay was 6.18, again between the values reported for galanthamine (pIC_{50} = 6.46) and physostigmine (pIC_{50} = 5.93).

Conclusions and Outlook

The preparation of chemically engineered extracts thorough the chemical alteration of reactive molecular fragments that are very common in natural products allows us to use known and unknown natural scaffolds for the generation of potentially bioactive derivatives. This can be envisaged as a way to recycle these scaffolds through directed decoration with unnatural functionalities.

The potential of CEEs to produce bioactive compounds depends in part on the power of numbers: the higher the proportion of altered components in the extracts is, the better are the chances of generating interesting compounds. In order to alter as many compounds as possible, it is necessary to identify and transform molecular fragments that are present in most natural components of the extracts. The CEEs reported to date have been prepared using reagents that focus on the transformation of only one high frequency group. The application of sequential reactions

directed to transform different common functionalities would increase the impact in composition. Our calculations indicate that transformation of any pair of the three high-frequency functional groups (carbonyl, −OH, double bonds) would lead to the alteration of more than 87% of the components of a hypothetical extract; and the transformation of all three would alter 97% of the components.

Aside from the proportion of modified compounds, the properties of the chemically engineered extracts will depend on the molecular fragments that are incorporated in the process. Initial reports describe hydrolysis or incorporation of molecular moieties that are uncommon in natural products (arylsulfonyl radical, halogens, etc). However, further developments could incorporate pharmacophores to direct the changes in biological properties toward the desired bioactivities.

Key for the preparation of CEEs is the methodology used for functional group transformation. The chemistry involved needs to be compatible with complex mixtures. Since the exact composition of the starting material is unknown, reagents are used in excess. However, the excess of reagents needs to be removed from the CEEs to avoid interferences with biological screening. The development of simple synthetic methodology in solution and on solid phase will be instrumental in the preparation of diverse CEEs.

Support of this work by grants from the Agencia Nacional de Promoción Científica y Tecnológica (ANPCyT, PICT 2006 995) and Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET) is gratefully acknowledged. I.A.R, M.O.S., and L.M. are supported by CONICET fellowships. We acknowledge especially a dedicated group of co-workers whose names and contributions are referenced.

Note Added after ASAP Publication. This paper was published on the Web on February 28, 2011 with an incorrect citation for reference 40. The corrected version was reposted on March 17, 2011.

BIOGRAPHICAL INFORMATION

I. Ayelen Ramallo completed her Ph.D. in organic chemistry at the University of Rosario in 2010 with Prof. Ricardo L. E. Furlan. Currently, she is carrying out postdoctoral research in the analytical chemistry field with Prof. Gabriela Ibañez and Prof. Alejandro Olivieri, supported by a CONICET fellowship.

Mario O. Salazar was born in Balcarce, Argentina, in 1981. He graduated in Pharmacy (2005) and received a Ph.D. in organic chemistry (2010) at the University of Rosario under the supervision of Ricardo L. E. Furlan. Currently, he is working in the NMR analysis of mixtures as part of his postdoctoral work with Manuel González Sierra at the University of Rosario.

Luciana Méndez was born in 1980 in Rosario, Argentina. She completed her Ph.D. in solid phase organic synthesis in 2009 at the Rosario Chemistry Institute (IQUIR-CONICET) under the supervision of Professor Ernesto Mata. As part of her postdoctoral research, she is currently working in halogenation reactions of natural extracts with Professor Ricardo Furlan.

Ricardo L. E. Furlan completed his Ph.D. in organic chemistry at the Rosario Chemistry Institute in 1999 with Prof. O. A. Mascaretti and carried out postdoctoral research with Prof. J. K. M. Sanders at the University of Cambridge. Since 2002, he is an Assistant Professor at the University of Rosario and a researcher of the National Council of Research. His research interests lie in the study of different strategies to generate molecular diversity including dynamic combinatorial chemistry and chemically engineered extracts.

FOOTNOTES

*To whom correspondence should be addressed. E-mail: rfurlan@fbioyf.unr.edu.ar. Tel/Fax: +54 341 4375315.

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