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A pilot study of urinary peptides as biomarkers for intelligence in old age

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ABSTRACT

Intelligence is an important indicator of physical, mental and social well-being. In old age, intelligence is also associated with a higher quality of life and better health. Heritability studies have shown that there are strong genetic influences, yet unknown, on intelligence, including in old age. Other approaches may be useful to investigate the biological foundations of intelligence differences. Proteomics is a proven technique in revealing biomarkers for certain illnesses. In this pilot study, forty individuals were selected as the cognitive extremes from over 750 people in the Lothian Birth Cohort 1936 (age ~72 years) based on their high and low intelligence scores, as measured by a general cognitive ability factor. Urine samples were used as a stable, reliable and abundant source of proteins. Using capillary electrophoresis coupled to mass spectrometry (CE-MS) technology, the proteome of the high and low intelligence groups was determined. Data were calibrated and matched against the human urinary database, to enable comparative assessment. At a nominal significance level ($P<0.05$), there were several candidate proteins for association with intelligence, including a zinc finger protein (ZNF653) that has been associated with cognitive deficits, and complement C3 and collagen fragments that have been associated with Alzheimer's disease. Results are preliminary, do not survive multiple testing correction, and require validation. This pilot study shows the potential of this novel proteomics approach, and its applicability to understanding the biological foundations of intelligence differences.

1. Introduction

Higher intelligence is associated with relative success in education, better performance in the workplace, and better physical and mental health and a longer life (Batty et al., 2007; Deary, Strand, et al., 2007; Gottfredson, 1997). In old age, maintaining cognitive ability is associated with a higher quality of life and being able to live independently. This is especially important, because society has an increasing proportion and absolute number of older people. It is a high research priority to discover the determinants of cognitive ability differences in old age (Deary, Johnson, & Houlihan, 2009). One appropriate place to search for a proportion of these differences is in the genes. The heritability (additive genetic contribution) of intelligence is about 50% across all studies, but it increases substantially from childhood to adulthood (Deary, Johnson, & Houlihan, 2009). It is still highly heritable, probably at least 60%, in old age. Despite

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numerous studies, no single genetic locus has been associated definitively with variance in cognitive ability, with the possible exception of a contribution of about 1% of the variance from *APOE* in old age. Considering the high heritability of intelligence this may seem surprising. However, cognitive ability is likely to be under the influence of many genes each having a very small effect, and the specific genetic variants may be located outside the coding regions of the genes making them harder to identify (Deary, Johnson, & Houlihan, 2009).

The dearth of positive results to date from molecular genetic studies of cognitive abilities means that there are few clues from genetic studies to the mechanistic biology of cognitive differences, including in old age (Deary, Penke, & Johnson, 2010). Searches for other biomarkers of cognitive differences and age-related cognitive changes have used blood-derived samples and examined, for example, inflammation biology (e.g. C-reactive protein; Luciano et al., 2009), telomeres (e.g., Harris et al., 2006), oxidative stress genes (e.g., Harris et al., 2007), and cortisol (and the hypothalamic-pituitary-adrenal axis more generally; e.g., MacLullich et al., 2005). Whereas there are some positive leads from these types of investigations, the effect sizes are typically small, the results often unreplicated, and there are indications of possible reverse causation; i.e., cases where cognitive traits appears to result in biomarker levels rather than the reverse (e.g. Luciano et al., 2009). In summary, neither the molecular genetic approach nor the search for cognitive biomarkers has, as yet, provided much insight into the mechanistic biology of cognitive differences though the lifecourse. A review of possible plasma biomarkers of mild cognitive impairment and Alzheimer's disease suggested some that might be useful to test in normal cognitive ageing, but they are likely to suffer from the same limitations as previous studies (Song et al., 2009).

The approaches described in the previous paragraph are hypothesis-driven. However, technical advances in mass spectrometry have brought about the possibility of a complementary, hypothesis-free, multi-parametric "omic" approach to biomarker discovery in intelligence. This alternative approach is to look directly at the abundance of the proteins under the control of the multiple genetic variants that are thought to influence intelligence differences. Moreover, proteomics reflects epigenetic influences on gene expression. Therefore, a proteomic approach is a logical and potentially fruitful way to study differences in intelligence–as suggested by, for example, Plomin et al. (2006)–as it has been for many other age-related disorders and traits (Schiffer et al., 2009). We propose to use urine as an easily collectable source of proteins. Urine represents an excellent specimen for proteome analysis, as it can be obtained in high quantities without the need for special collection procedures (Thongboonkerd, 2007), shows higher stability than blood (Kolch et al., 2005; von Zur Muhlen et al., 2009), and enables the identification of valid biomarkers for renal as well as systemic diseases (Decramer et al., 2006; Weissinger et al., 2007). Mass spectrometry analysis of the human urinary proteome has identified >1500 proteins in the soluble fraction (Adachi et al., 2006) and >1100 proteins in urinary exosomes, 177 of which were represented in the OMIM database of disease related genes (Gonzales et al., 2009). Recently, we identified and validated urinary proteomic biomarkers for diabetes, diabetes-associated micro- and macrovascular complications (Meier et al., 2005; Mischak et al., 2004; Rossing, Mischak, Dakna, et al., 2008; Rossing, Mischak, Rossing, et al., 2008; Snell-Bergeon et al., 2009; von zur Muhlen et al., 2009; Zimmerli et al., 2008), and ageing (Zürbig et al., 2009).

Proteome analysis, therefore, holds the promise of delivering some insights into the pathophysiological changes associated with several bodily disorders and complex quantitative traits, including cognitive abilities. Encouraged by these findings and the success of pilot studies in other research areas (Airoldi et al., 2009; Roesch-Ely et al., 2010), we investigated whether intelligence in old age, and the changes in extracellular matrix associated with it, can be assessed in part by urinary biomarkers.

2. Methods

2.1. Participants, urine collection, and the selection of cognitive extremes

We undertook a pilot study in 40 adults from the Lothian Birth Cohort 1936 (LBC1936) (Deary, Gow, et al., 2007) aimed at assessing the ability of mass spectrometry to identity urinary protein biomarkers for cognitive ability in the general older population. All individuals in the LBC1936 were born in 1936 and attended school in Scotland in 1947. At an average age of 11 years they took a valid IQ-type test–a version of the Moray House Test no. 12 (MHT)–in the nationwide Scottish Mental Survey 1947 (SMS1947; $N = 70{,}805$) (Deary, Whalley, & Starr, 2009). The LBC1936 were first examined by our team between 2004 and 2007 at a mean age of 70 years (Deary, Gow, et al., 2007). They re-sat the same mental test that they took at age 11, and underwent other cognitive and medical tests, as described elsewhere in detail (Deary, Gow, et al., 2007; Deary, Strand, et al., 2007). At age ~72 years, the surviving, available, and interested LBC1936 participants were recruited a second time, as still relatively healthy participants of the SMS1947 who were mostly living in the Edinburgh area (Lothian) of Scotland. All participants in the study lived independently in the community and travelled to the Wellcome Trust Clinical Research Facility (WTCRF) at the Western General Hospital, Edinburgh, UK, for testing. The local ethics committees approved the study, and all subjects gave informed consent. The study adhered to the Helsinki Declaration.

Urine samples were available for 762 (397 males) participants. The participants were asked to provide a first-pass midstream urine specimen from first thing in the morning. The urine samples were separated into three 1ml aliquots and stored at −40 °C. Participants were excluded based on the following criteria; any history of myeloma, diabetes, renal disease, kidney disease, bladder disease, prostate disease; evidence of a current infection defined by C-reactive protein >10 mg/L; hemoglobin <130 g/l; history of hypertension; body mass index <20 or >30 kg/m^2; and estimated glomerular filtration rate (eGFR) <60 ml/min. Samples were included if the urinary albumin/creatinine ratio was <2.5 mg/mmol (men) or <3.5 mg/mmol (women).

Because this was a pilot study, in order to maximise group differences in the cognitive phenotype, we chose 40 samples,

representing one or other extreme trait level on a general cognitive ability factor. Twenty samples in the high and low cognitive trait groups were selected, with ten males and females in each. A general cognitive ability factor (*g* factor) was derived from the whole sample (*n* with relevant data = 778) from a principal components analysis of six Wechlser Adult Intelligence Scale-III^{UK} (WAIS-III^{UK}; Wechsler, 1998) subtests: Backward Digit Span, Letter–Number Sequencing, Matrix Reasoning, Block Design, Digit Symbol, and Symbol Search. These were performed during the second recruitment wave when the subjects were aged ~72 years old [Mean *g* factor score = 0, Standard deviation (SD) = 1]. Regression scores were calculated for the first unrotated principal component with SPSS 14.0 for Windows.

The sample size used in the analyses was 40 individuals (20 females and 20 males). The mean age in the higher cognitive group was 72.25 years (standard deviation = 0.67 years) and was 72.36 years (standard deviation = 0.55 years) in the lower cognitive ability group. There were no biochemical abnormalities detected. In all samples, the eGFR was >60 ml/min and the mean albumin/creatinine ratio was <2.5 mg/mmol in men and <3.5 mg/mmol in women, demonstrating no detectable kidney problems (eGFR measuring the excretory function of the kidneys and albumin/creatinine ratio measuring albumin protein "leaking" into the urine). The mean general cognitive ability score in the higher intelligence group was 1.84 (SD = 0.56) and was -1.36 (SD = 0.44) in the lower intelligence group.

2.2. Capillary electrophoresis coupled to mass spectrometry (CE-MS) analysis

A 0.7 mL urine aliquot was thawed and diluted with 0.7 mL 2 M urea, 10 mM NH_4OH containing 0.02% SDS. Proteins >20 kDaltons (Da) were removed by ultracentrifugation through Centrisart filters (Sartorius, Goettingen, Germany) at 2300*g* relative centrifugal force (rcf) until 1.1 ml filtrate was obtained. The filtrate was desalted on a PD-10 column (GE Healthcare, Sweden) equilibrated in 0.01% aqueous NH_4OH (Roth, Germany). Samples were lyophilized and stored at 4 °C, until resuspension in High Performance Liquid Chromatography (HPLC)-grade H_20 to a final protein concentration of 0.8 μg/μL prior to analysis. Analysis and data processing were performed in accordance with current guidelines for clinical proteome analysis (Mischak, Apweiler et al., 2007; Taylor et al., 2007) using a P/ACE™ MDQ capillary electrophoresis system (Beckman Coulter, Fullerton, USA) on-line coupled to a Micro-Time-of-flight mass spectrometry (TOF MS) (Bruker Daltonic, Bremen, Germany) (Kolch, Neususs, Peizing, & Mischak, 2005; Theodorescu et al., 2005). Spectra were accumulated every 3 s over a mass-to-charge ratio (m/z)-range of 350–3000 Da. Details on accuracy, precision, selectivity, sensitivity, reproducibility, and stability of the CE-MS method have been reported (Theodorescu et al., 2005).

2.3. Data processing

Mass spectral ion peaks representing identical molecules at different charge states were deconvoluted into single masses using MosaiquesVisu (Wittke et al., 2003). The software employs probabilistic clustering and uses both isotopic distribution and conjugated masses for charge-state determination of peptides/proteins. The resulting peak list characterizes each polypeptide by its molecular mass, CE-migration time, and ion signal intensity (amplitude). Data were normalized using a calibration method for CE-migration time and ion signal intensity on the basis of 'internal standard' peptides, which we proved to be superior over creatinine normalization (Jantos-Siwy et al., 2009; Theodorescu et al., 2006). All peptides were deposited, matched, and annotated in a Microsoft SQL database. Peptides were considered identical when mass deviation was lower than 50 ppm for small peptides or 75 ppm for larger peptides and proteins. Deviation of migration time was below 0.35 min.

2.4. Generation of polypeptide pattern

A polypeptide biomarker model was generated by combination of peptides that were differentially distributed between the two groups separated by high and low *g* factor scores using the support-vector-machine (SVM)-based MosaCluster software (Theodorescu et al., 2006). In SVM, a sample is regarded as a p-dimensional vector with p being the number of peptides included in the pattern. The algorithm constructs a (p-1)-dimensional separation plane between the high and low *g* factor vectors. From all possible hyperplanes that separate the high and low *g* factor groups, the one with the largest distance to the nearest data points on both sides is selected. Classification is performed by determining the Euclidian distance of the data point to the maximal margin of the hyperplane and assignment of a positive or negative value depending on which side of the hyperplane, high or low *g* factor, the vector is located.

2.5. Sequencing of peptides

Sequences were obtained and validated as described in detail for the standard urine sample (Mischak et al., 2010). Briefly, peptides were separated by nanoflow reversed phase (nRP)-HPLC (Agilent 1100; flow split by tee to ~60 nL/min) and introduced into an Electron Transfer Dissociation-capable Finnigan linear trap quadruploe (LTQ) quadrupole linear ion trap via nanoelectrospray ionization (nESI) as previously described (Good et al., 2010). The resulting MS/MS data were submitted to MASCOT (www.matrixscience.com) for a search against human entries in the Swiss-Prot database without any enzyme specificity with an accepted parent ion mass deviation of 10 ppm and an accepted fragment ion mass deviation of 500 ppm. Only search results with a MASCOT peptide score equally or higher as the MASCOT score threshold were included. An additional search was employed using OMSSA (http://pubchem.ncbi.nlm.nih.gov/omssa/).

2.6. Statistical analysis

Sensitivity, specificity, 95% confidence interval (CI) and area under the curve (AUC) were calculated based on receiver operating characteristic (ROC) analysis; MedCalc version 9.5.2.0 (MedCalc Software; Mariakerke, Belgium). For identification of potential biomarkers, the Wilcoxon *T* test was applied because the data are not normally distributed. *P*-values were calculated using the log transformed intensities. The

conventional *P*-value <0.05 was applied first. Then, to adjust for multiple testing, the false discovery rate was controlled at 0.05 according to Benjamini & Hochberg (1995).

3. Results

All 40 urine samples from all subjects included in the study were analyzed using CE-MS and evaluate peptides deposited in the database, 3664 could be detected. 1309 peptides passed the threshold of being present in >30% in at least one of the two groups and were investigated. Data from the subjects were grouped according to the general cognitive (*g*) factor, high or low, and examined for significant differences. A graphic depiction of the data is shown in Fig. 1a. The peptides are shown as a 3-dimensional plot, mass versus migration time, and the peak height reflects signal intensity. Some apparent differences between the datasets appear visible. No significant biomarkers withstood adjustment for multiple testing (all adjusted *P*-values >0.1, data not shown). Without adjustment for multiple testing, several potentially significant peptides (*P*-value <0.05) can be identified. The results of the statistical comparison are given in Supplementary Table 1 and the identified peptides are shown in Table 1. In Fig. 1b, the distribution of the 75 potential biomarkers is shown. These peptides show highly different distributions in the two groups.

We investigated the value and validity of the potential biomarkers further, based on the data available. We employed the 75 most significant candidates in an SVM-driven model, and assessed the model using complete take-one-out

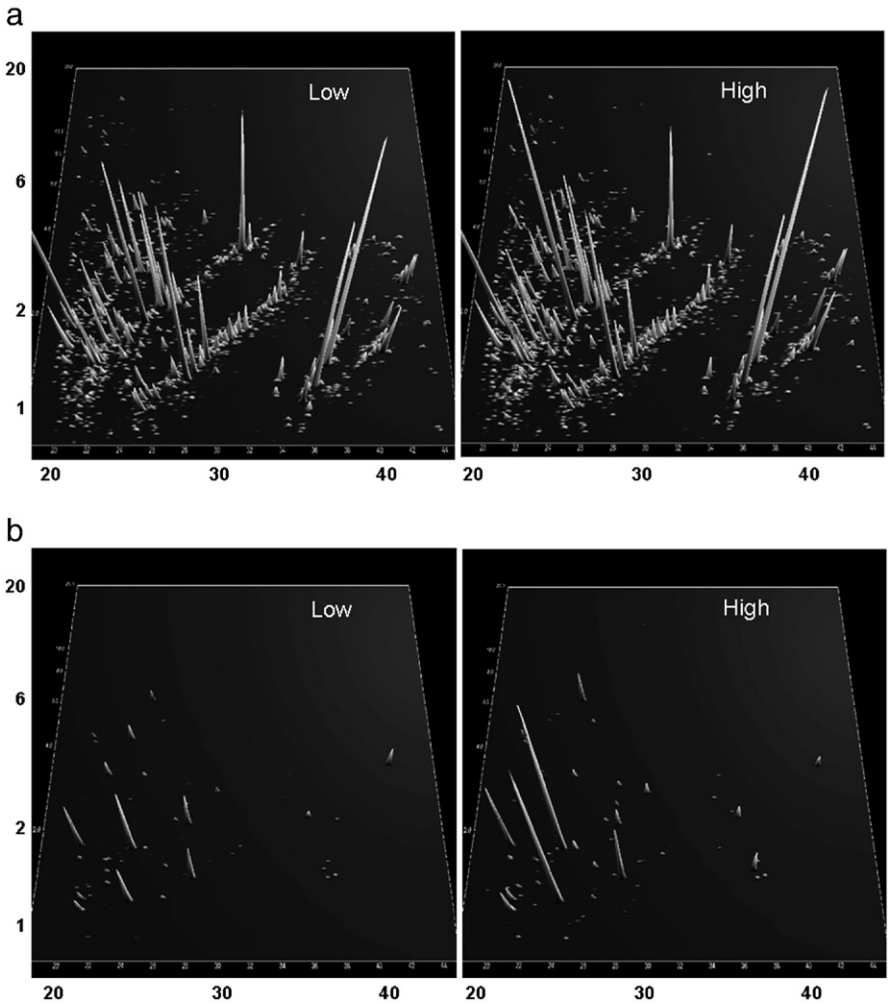

Fig. 1. Compiled protein patterns of the capillary electrophoresis coupled to mass spectrometry (CE-MS) analysis of urine samples from the Lothian Birth Cohort 1936 (LBC1936) groups of higher intelligence ($N=20$) and lower intelligence ($N=20$) examined in this study. The molecular mass on a logarithmic scale (0.8–20 kDa, indicated on the left) is plotted against normalized migration time (18–45 min, indicated on the bottom). Signal intensity is encoded by peak height and shade of grey colour. Subtle differences in peptides present between apparently high and low intelligence in LBC1936 can be identified (a). Panel b shows the distribution of only the 75 peptides that revealed the largest differences between the two groups (unadjusted *P*-value <0.05). To better visualize the potential biomarkers, peak intensity is enlarged 5-fold in panel b, in comparison to panel a. When examining only these 75 potential biomarkers, differences between high and low intelligence in LBC1936 appear quite evident.

Table 1
Protein fragments identified from 75 potential biomarkers for the discriminatory model between individuals with high and low intelligence in the Lothian Birth Cohort 1936 are listed. The peptide identification number in the dataset (Coon et al., 2008), the original protein name, and the sequence of the peptide in the respective protein sequence are given.

Protein identification number	Protein name	Sequence
59022	Uromodulin	SVIDQSRVLNLGPITR
108021	Complement C3	EGVQKEDIPPADLSDQVPDTESETR
124479	Collagen alpha-1 (I) chain	PpGESGREGAPGAEGSPGRDGSpGAKGDRGETGP
94807	Collagen alpha-1 (I) chain	AGQDGRpGPpGppGARGQAGVmGFpG
99691	Zinc finger protein 653	PEAEAEAEAGAGGEAAAEEGAAGRKARG
51175	Collagen alpha-1 (I) chain	EGSpGRDGSpGAKGDRG
79786	Collagen alpha-1 (I) chain	ADGQPGAKGEPGDAGAKGDAGPpGP
77018	Collagen alpha-1 (I) chain	DGQPGAKGEpGDAGAKGDAGPPGp

cross-validation, resulting in an AUC of 0.975, 100% sensitivity and 85% specificity (P-value <0.0001). The ROC curve is shown in Fig. 2.

As shown in Supplementary Table 1, we were able to obtain the sequences from some peptides that may be differently distributed between the two groups. When examining the peptides that appear to be of the highest significance, we found a >2-fold increase of a C-terminal peptide from uromodulin (see Fig. 3), the most abundant protein in normal human urine, in the group with the high g score. A peptide from Complement C3 C-terminus and four peptides derived from Collagen alpha-1 (I) chain were found substantially increased in the urine samples of the subjects with high intelligence. The N-terminus of the Zinc finger protein 653, a transcriptional repressor, was increased 7-fold in the individuals with a low g score, as was a peptide derived from Collagen alpha-1 (I) chain (see Fig. 3).

These results indicate that urinary proteomic biomarkers for cognition may exist, but the statistical power of the pilot study is not sufficient to confirm this assumption. Estimates on the sample size required, based on resampling of data presented here and applying the same adjustment for multiple testing using the false discovery rate, indicate that approximately 200 high- and low-scorers each need to be analyzed to enable identification of 75 biomarkers with good confidence (q-value <0.01), and 300 cases and controls would need to be analyzed to enable identification of 90 biomarkers.

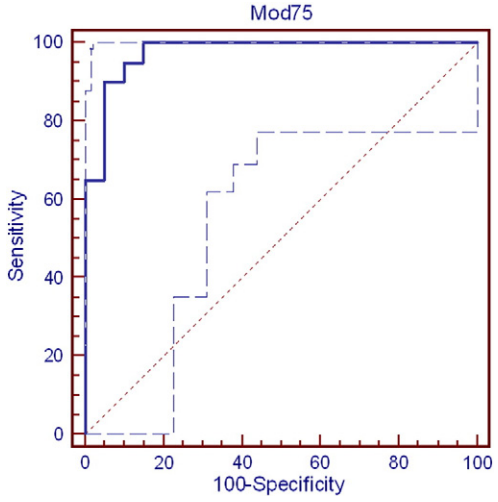

Fig. 2. Performance of the proteomic biomarker model based on 75 urinary peptides that may be biomarkers for cognition in old age. Shown is the receiver operating characteristic (ROC) analysis of the classification results obtained differences between the Lothian Birth Cohort 1936 (LBC1936) group of higher intelligence ($N=20$) and lower intelligence ($N=20$) after complete take-one-out cross-validation. The numerical values of the classification obtained employing the support-vector-machine (SVM)-driven model based on the 75 biomarkers was examined. The 95% confidence interval is indicated by the dashed line.

4. Discussion

The data indicate that potential differences between older people of high and low general intelligence can be identified in the proteins in human urine. There are certain reasons to believe that the four proteins suggested in this study may be valid and worthy of replication attempts. Three of the four proteins that gave rise to the peptides with the highest significance have functional evidence to prioritise them for follow-up. First, uromodulin is coded for by the gene *UMOD* and defects in this gene cause renal disorders, characterised by hyperuricemia, gout, and progressive renal failure. Genetic variants associated with gout and serum uric urate levels have been associated with memory performance in human studies (Houlihan, Wyatt, et al., 2010). Second, the gene encoding Complement C3, *C3*, has been associated with Alzheimer's disease (Giedraitis et al., 2009). The third candidate, ZNF653, is a zinc finger protein. Whereas there is no evidence to suggest this protein specifically, zinc finger proteins have been associated with cognitive deficits in certain illnesses such as X-linked mental retardation (Shoichet et al., 2003), schizophrenia (Walters et al., 2010), and autism (Willemsen et al., 2010). Recently, several reports indicated an increase of collagen, or collagen fragments that are generated during the assembly of collagen (indicating increased collagen deposit), being associated with Alzheimer's disease and mild cognitive impairment (Luckhaus et al., 2009; Tong, Xu, Scearce-Levie, Ptacek, & Fu, 2010). These reports support the hypothesis that the upregulation of the two fragments of collagen alpha-1 we report here, which are degradation products of the mature collagen fibres, indicating a reduction of collagen fibre in the tissue, is associated significantly with advanced intelligence or cognitive function.

The study has a number of strengths. First, it opens a new avenue of research on cognitive differences, one that is not prey to the highly polygenic influences on cognitive differences. Thus, the protein levels capture the many regulatory influences on any one gene transcribing a given protein.

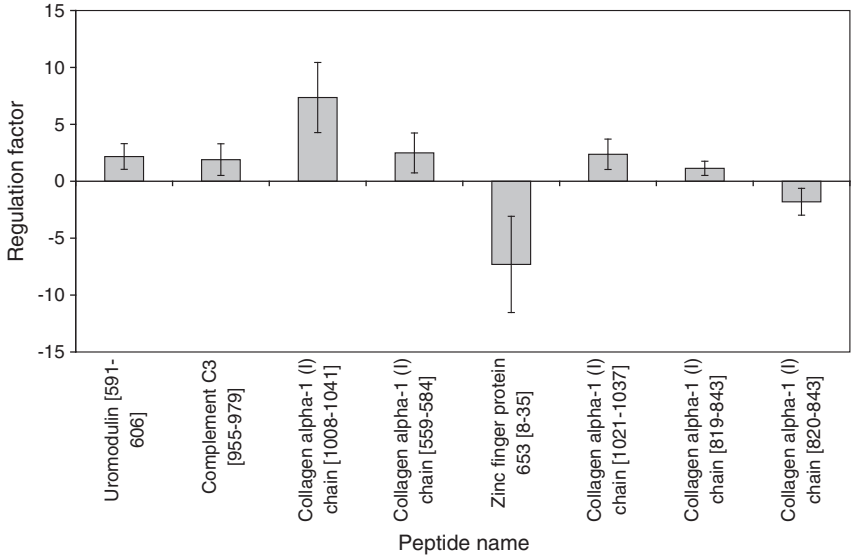

Fig. 3. Regulation of the sequenced peptide markers in urine samples of the LBC1936 cohort. Displayed is the regulation of an uromodulin fragment, a Complement C3 fragment, five collagen alpha-1 (I) fragments, and a Zinc finger protein 653 fragment. The regulation is the fold-difference of mean signal intensity between high and low intelligence groups. The errors bars represent the standard deviation. The bar-chart is ordered according to significance value from left to right.

Second, we deliberately chose extreme cognitive scores form the sample, based upon multiple tests from one of the best validated intelligence batteries. Third, there was a conservative approach to significance testing. However, because the study was novel, the top proteins were still named, even though they did not meet rigorous adjustments for multiple testing. Finally, the results from the complete take-one-out cross-validation suggest that validation of our preliminary results of the biomarker model in an independent cohort could be successful. In order to establish validity of the differences between higher and lower intelligence, a substantially larger number of additional samples needs to be analyzed. Based on the calculations and also on experience in other studies that clearly indicate a significant benefit of a larger number of biomarkers in a biomarker model, approximately 200 higher- and lower-scorers on intelligence tests should suffice to identify >75 biomarkers, which would enable establishment of a stable classifier. This classifier would have to be validated in an independent set of samples (according to guidelines just published, Mischak et al. STM 2010); consequently, a total of 600 independent samples would be required for a study to prove the concept that urinary biomarkers can be employed in the unbiased assessment of cognitive skills. This is planned in future work.

The study also has limitations. First, the main limitation is the small number of subjects in each group and the consequent low power. This was partly because the cost and labour involved in proteomic analysis is still relatively large. However, we consider the study to be useful in establishing feasibility and encouraging this approach among others. Considering the fact that the complete take-one-out cross-validation still allowed almost perfect classification accuracy (1/40 was incorrectly assigned) indicates that the combination of biomarkers chosen in the model consisting of 75 urinary peptides is associated with cognitive function, and that these preliminary findings warrant further investigation.

Second, the large number of outcome variables and the small number of subjects, means that type 1 and 2 statistical errors must be expected. This is illustrated in a possible artefactual result where four of the five peptides from the collagen 1 fragments are consistently upregulated in the group with the higher *g* score except for Collagen alpha-1 chain (820–843) which is upregulated in the group with the low *g* score. This experimental artefact may be due to the low statistical power in this small cohort, which will be resolved when more samples are analyzed. Thus, this observation should not be interpreted with confidence.

A third limitation is the study sample. The subjects in the "lower" cognitive group were not especially low, because the LBC1936 cohort is, overall, cognitively quite advantaged (Deary, Gow, et al., 2007). Furthermore, the proteomic analysis was based on urine which, of course, does not contain all proteins that might usefully be assayed. In particular, proteomic profiles reflecting genes expressed where their end-products are excreted in urine may differ from those in the brain; ante-mortem brain tissue is very rarely available for proteomic analysis and post-mortem tissue proteomics may be substantially influenced by agonal events. Nevertheless, urinary proteomics has been useful in providing biomarkers for other conditions and traits and given limited access to brain tissue, it is worthy of further investigation with regard to intelligence traits.

There were further technical limitations to the study. First, there is a lack of sequence for several of peptides that appear to be differentially distributed. As outlined in detail (Mischak et al., 2009), sequencing of naturally occurring peptides represents substantial challenges, among other due to the potential of unknown posttranslational modifications being present. However, the peptides described here are characterized in sufficient detail (by accurate mass, migration time in CE, and their presence in a significant number of urine samples) that allows allocating identity with very high

confidence (see also Coon, et al., 2008). This will potentially enable sequencing those peptides in the future by using substantially more sample, and/or other, better suited mass spectrometers. Second, few peptides could be identified by sequencing, but was hindered by several obstacles associated with MS sequencing of naturally occurring peptides; tryptic digests cannot be utilized due to a loss of connectivity to the original identification parameters (Chalmers et al., 2005). Major obstacles are suboptimal employment of proteomic search machines (like Mascot or OMSSA) for naturally occurring peptides (Fliser et al., 2007; Mischak, Julian, & Novak, 2007), as well as the chemical nature of the peptides that prevents successful sequencing (Zürbig et al., 2006).

It is hoped that, among other "omic" approaches to identifying the mechanisms underlying cognitive ability and its changes through the human lifecourse, proteomics will be added. Urine samples are especially easy to collect by comparison with most other biological samples, and so any emerging markers derived from urine could offer a convenient test for cognitive level and changes. Future studies should have larger samples that are well-characterised on cognitive abilities. With the relative lack of information currently available from other biological approaches to cognitive differences, proteomics has the advantage of assessing the biological end points of many influences on protein synthesis and breakdown, and so might provide a relatively fruitful source of cognitive biomarkers. The identification of such biomarkers should provide us with a deeper understanding of the pathophysiological changes associated with the various degrees of cognitive decline in old age, and may even result in advancements in therapeutic strategies by early detection of dementia.

Supplementary materials related to this article can be found online at doi:10.1016/j.intell.2010.11.001.

Disclosures

H Mischak is founder and co-owner of Mosaiques diagnostics GmbH, which developed the CE-MS technology and the MosaiquesVisu software. P Zürbig is an employee of Mosaiques diagnostics GmbH.

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