Grant Impact Report

Name	Holger W. Auner, MD PhD
Job title	Clinical Senior Lecturer and Honorary Consultant in Haematology
Organisation	Imperial College London
Co-investigators (if applicable)	
Grant awarded	Early Stage Research Start-up Grant
Year awarded	2017
Date started	01/01/2017
Date completed	31/12/2017
Total amount expended (£)	10,000

This is the BSH grant impact report form. Please enter the full grant details above, and fill out the form below. The form should be completed electronically and sent to grants@b-s-h.org.uk. Please note that the report can only be accepted if all sections have been completed in full.

In addition: Please include a recent photo of yourself.

Your grant report and photo may be published in our communications materials, including our website and social media platforms.

To see previously published grant impact profiles, please visit our website.

1. Please summarise what the grant enabled you to achieve; what would not have been possible without the funding? (Up to 500 words)

My research group works on aspects of intracellular protein homeostasis (proteostasis) in cancer cells and non-transformed cells, with a particular interest in multiple myeloma. The ultimate goal of our work is to find ways to disrupt proteostasis in malignant cells while largely sparing non-cancerous cells. This aim builds on the proposition that cancer cells, and in particular myeloma cells, are highly dependent on the mechanisms that co-ordinate protein degradation and synthesis. This notion is supported by the clinical success of inhibitors of the proteasome, the main effector of intracellular protein degradation.

The BSH grant allowed us to complete a set of experiments that formed a key part of a project that has now been completed, with a manuscript under review (May 2018). The grant also enabled us to conduct a set of preliminary experiments that supported a successful grant application with an industry partner and another grant application that has been submitted to a UK organisation. While the experiments that were funded by the BSH grant formed part of a larger work programme, the results of these experiments became important components of both the submitted manuscript and the grant applications, providing critical support for our conclusions and hypotheses.

2. Briefly describe the aims and intended outcomes of this project. Please clearly indicate if there is any sensitive information in this report that should remain confidential for now. (Up to 300 words)

We previously identified inadequate fine-tuning of protein synthesis by the translation factor eIF2α, and a detrimental rearrangement of the intracellular amino acid pool, as important mechanism by which inhibitors of an ATPase that mediates protein degradation induce cancer cell death. We had also found preliminary evidence that led us to hypothesise that an amino acid-sensing kinase that regulates protein synthesis could be a potential therapeutic target.
Building on our advances, and tackling key clinical issues, we proposed to further investigate the following questions: (1) How does the eIF2α kinase *EIF2AK4* determine cancer cell fate after proteotoxic insults? (2) How does *EIF2AK4* drive the recovery of cancer cells from proteotoxic stress? (3) How can we exploit nutrient limitations to optimise treatments that target protein degradation? We proposed that these studies would provide important biological insights into cancer cell protein metabolism. Moreover, they could change clinical practise through improved therapeutic approaches that target proteostasis, or through identification of resistance biomarkers.

3. **Describe the key outcomes to date, including whether this grant has resulted in further research. Please summarise your conclusions. (Up to 600 words)**

The experiments that were supported by the BSH grant include metabolomic studies (GC-MS) on cancer cells that were (1) maintained in nutrient-limiting conditions, (2) treated with a protein degradation inhibitor, and (3) in which *EIF2AK4* was depleted by shRNAs. We also analysed specific candidate signalling pathways (such as ERK), alternative protein degradation processes (macro-autophagy) and overall protein synthesis (by immunoblotting for puromycinylated proteins). The results, which are still unpublished and cannot therefore be presented in detail, support the hypotheses outlines in the previous section. We will acknowledge BSH support in all relevant publications and will continue to update BSH on the progress of funding applications that were supported by the award.

4. **List published papers, oral and/or poster presentations as a result of this grant.**
Include manuscripts in preparation or in submission / under review, prefaced by an asterisk.

*One manuscript currently under review, further information to be included once published.

5. **Did any patent applications arise from this work? (If yes, please detail. Up to 200 words)**

NO (but these may still arise).

6. Were you successful in any further grant applications as a result of this work? (If yes, please detail. Up to 200 words) YES (please see section 1)
7. Did new collaborations arise from this work? (If yes, please detail. Up to 400 words) YES. The work has contributed to the establishment of collaborations with several investigators from the Department of Mathematics at Imperial College London and with Dr Martin Kaiser (ICR).
8. What was the funding amount you received and how was it actually spent? (detail item/activity and amount spent in pounds) *Please see attached excel spreadsheet.*

9. What are the future research priorities in this area?

- Validating EIF2AK4 as a target for anticancer therapy
- Establishing the mechanisms that govern proteaome inhibitor resistance
- Further development of VCP/p97 inhibitors in early-phase clinical trials
- Elucidating the interplay between micro-environmental nutrient shortages and adaptive mechanisms activated by proteasome inhibitors

Transaction Supercategory	GL Period	Cost (GBP)	Employee/Supplier	Expenditure Comment
CONSUMABLES	Mar-17	98.90	Sigma-Aldrich Company Ltd	EASYFLASK(TM) 175 CM2, V/C CAP,
CONSUMABLES	Mar-17	36.96	Fisher Scientific Ltd	2.5LT Ethanol absolute 99.8+%, C
CONSUMABLES	Mar-17	240.00	Life Technologies Limited	FBS HEAT INACT. S.AMERICAN(CE)
CONSUMABLES	Mar-17	216.88	Life Technologies Limited	FBS HEAT INACT. S.AMERICAN(CE)
CONSUMABLES	Mar-17	77.34	Sigma-Aldrich Company Ltd	3-METHYLADENINE
CONSUMABLES	Mar-17	134.50	Sigma-Aldrich Company Ltd	BAFILOMYCIN A1 READY MADE SOLUTI
CONSUMABLES	Mar-17	86.70	Sigma-Aldrich Company Ltd	U0126 MONOETHANOLATE, >/=98% (HP
CONSUMABLES	Apr-17	152.70	Starlab (UK) Ltd	1000microl XL Graduated TipOne Filter Tip (Sterile), Racked
CONSUMABLES	Apr-17	169.00	Starlab (UK) Ltd	1250microl XL Graduated TipOne Tip, Refill
CONSUMABLES	Apr-17	53.20	VWR International Ltd	TUBE SECURE LOCK NATURAL PP 1.5M
CONSUMABLES	Apr-17	89.50	VWR International Ltd	FERROSTATIN-1
CONSUMABLES	Apr-17	89.50	VWR International Ltd	SURCHARGE
CONSUMABLES	Apr-17	-72.55	VWR International Ltd	SURCHARGE
CONSUMABLES	Apr-17	11.97	Fisher Scientific Ltd	2.5LT Methanol, extra pure, SLR.
CONSUMABLES	Apr-17	73.86	Scientific Laboratory Supplies Ltd	Hepes solution bioxtra 1 m ph 7.
CONSUMABLES	May-17	46.38	Starlab (UK) Ltd	0.2ml 8-Strip PCR Tubes, Individually Attached Flat Caps (X-Clear)
CONSUMABLES	May-17	28.50	VWR International Ltd	TEST TUBE 5ML PS ROUND BOTTOM 12
CONSUMABLES	May-17	155.54	Life Technologies Limited	MEM ALPHA W/O RIBOS W/GLUMAX-I
CONSUMABLES	May-17	147.19	VWR International Ltd	REAGENT WESTERN BLOT ECL 4000CMÃ
CONSUMABLES	May-17	382.00	VWR International Ltd	TUBE OPEN ULTRA
CONSUMABLES	May-17	-240.00	Life Technologies Limited	FBS HEAT INACT. S.AMERICAN(CE)
CONSUMABLES	May-17	-216.88	Life Technologies Limited	FBS HEAT INACT. S.AMERICAN(CE)
CONSUMABLES	May-17	240.00	Life Technologies Limited	FBS/reserve 3726119/lot 0170K08Q
CONSUMABLES	May-17	99.90	Fisher Scientific Ltd	250 UG O'GENERULER(TM) 1 KB PLUS
CONSUMABLES	May-17	26.69	Life Technologies Limited	ATTUNE WASH SOLUTION
CONSUMABLES	May-17	49.68	Sigma-Aldrich Company Ltd	TRYPAN BLUE SOLUTION, FOR MICROS
CONSUMABLES	May-17	61.20	VWR International Ltd	AZD6244
CONSUMABLES	May-17	57.50	Sigma-Aldrich Company Ltd	REFERENCE DYE FOR QUANTITATIVE P
CONSUMABLES	May-17	217.00	Life Technologies Limited	Top up of PO so invoice can be paid
CONSUMABLES	May-17	-0.12	Life Technologies Limited	Top up of PO so invoice can be paid
CONSUMABLES	May-17	40.80	New England Biolabs	Tris Buffered Saline (TBS-10X)
CONSUMABLES	May-17	105.60	New England Biolabs	Anti-rabbit IgG HRP-linked Antib
CONSUMABLES	May-17	28.79	Scientific Laboratory Supplies Ltd	PO 3609680 top up
CONSUMABLES	May-17	18.97	Sigma-Aldrich Company Ltd	D-(+)-GLUCOSE SOLUTION, 100 G/L
CONSUMABLES	May-17	155.00	Stratech Scientific Ltd	torin2 10mM in 1ml DMSO
CONSUMABLES	Jun-17	147.80	Starlab (UK) Ltd	pipette clinic top up PO 3555773 Invoice number 92023993 Voucher number 2814572
CONSUMABLES	Jun-17	23.15	Appleton Woods Ltd	Aluminium foil 50cm x 90m
CONSUMABLES	Jun-17	2.16	Medical Research Council	PO Closed ; PO: 3574454 ; Supplier: Medical Research Council ; Invoice: MRC_23881 ; Cost Centre:. WMEH
CONSUMABLES	Jun-17	12.96	Medical Research Council	sequencing
CONSUMABLES	Jun-17	-2.16	Medical Research Council	sequencing
CONSUMABLES	Jun-17	19.76	Bio-Rad Laboratories Ltd	INNER GLASS PLATE,5,M-P3
CONSUMABLES	Jun-17	71.75	Life Technologies Limited	96-WELL FAST THERMAL CYCLING
CONSUMABLES	Jun-17	276.50	Starlab (UK) Ltd	PO 3555773 Top up
CONSUMABLES	Jun-17	144.00	Stratech Scientific Ltd	CB5083 5mg
CONSUMABLES	Jun-17	420.85	Life Technologies Limited	AF488 ANNEXIN KIT 250 ASSAY
CONSUMABLES	Jun-17	309.70	Life Technologies Limited	ALAMARBLUE, 100ML
CONSUMABLES	Jun-17	154.40	Scientific Laboratory Supplies Ltd	GE Healthcare ECL Prime Western
CONSUMABLES	Jun-17	134.50	Sigma-Aldrich Company Ltd	BAFILOMYCIN A1 READY MADE SOLUTI
CONSUMABLES	Jun-17	374.40	Sigma-Aldrich Company Ltd	SYBR(R) GREEN JUMPSTART TAQ READ
CONSUMABLES	Jun-17	81.20	VWR International Ltd	HYPERFILM ECL 18X24CM
CONSUMABLES	Jun-17	28.10	Life Technologies Limited	7-AAD VIABILITY STAIN SOLN
CONSUMABLES	Jun-17	265.00	Abcam Ltd	GCN2 (phospho T899) antibody [EPR2320Y]
CONSUMABLES	Jun-17	599.32	Fisher Scientific Ltd	250 RXN NUCLEIC ACID PURIFICATIO
CONSUMABLES	Jun-17	44.80	New England Biolabs	Cell Lysis Buffer (10X)
CONSUMABLES	Jun-17	2.00	Sigma-Aldrich Company Ltd	ATF F
CONSUMABLES	Jun-17	2.00	Sigma-Aldrich Company Ltd	ATF4 R
CONSUMABLES	Jul-17	60.00	Sarstedt Ltd	PCR plate covers
CONSUMABLES	Jul-17	-5.00	Sarstedt Ltd	PCR plate covers
CONSUMABLES	Jul-17	29.40	Greiner Bio-One Ltd	7ml Bijou Container, no label, s
CONSUMABLES	Jul-17	71.75	Life Technologies Limited	96-WELL FAST THERMAL CYCLING
CONSUMABLES	Jul-17	194.56	Life Technologies Limited	PHIRE TISSUE DIRECT PCR MASTER
CONSUMABLES	Jul-17	183.20	New England Biolabs	Beta-Tubulin Antibody
CONSUMABLES	Jul-17	40.80	New England Biolabs	Tris Buffered Saline (TBS-10X)
CONSUMABLES	Jul-17	205.20	Fisher Scientific Ltd	1mL ROX solution, 50Â µM
CONSUMABLES	Jul-17	51.30	Fisher Scientific Ltd	1mL ROX solution, 50Â µM
CONSUMABLES	Jul-17	40.67	Fisher Scientific Ltd	MODIFYING ENZYME, DNASE I, RNASE
CONSUMABLES	Jul-17	36.81	Life Technologies Limited	10X REACTBUF(+MGCL2)FOR DNASEI
CONSUMABLES	Jul-17	243.60	VWR International Ltd	HYPERFILM ECL 18X24CM
CONSUMABLES	Jul-17	46.00	Sigma-Aldrich Company Ltd	REFERENCE DYE FOR QUANTITATIVE P
CONSUMABLES	Jul-17	374.40	Sigma-Aldrich Company Ltd	SYBR(R) GREEN JUMPSTART TAQ READ
CONSUMABLES	Aug-17	44.08	Bio-Rad Laboratories Ltd	Mini-PROTEAN Comb 15W 1.5 mm 40
CONSUMABLES	Aug-17	50.82	Chondrex Inc	N/A
CONSUMABLES	Aug-17	8.34	Chondrex Inc	concentrating solution 50ml
CONSUMABLES	Aug-17	183.20	New England Biolabs	Beta-Tubulin Antibody
CONSUMABLES	Aug-17	250.00	Sigma-Aldrich Company Ltd	ANTI-PUROMYCIN ANTIBODY, CLONE 1
CONSUMABLES	Aug-17	-12.50	Sigma-Aldrich Company Ltd	ANTI-PUROMYCIN ANTIBODY, CLONE 1
CONSUMABLES	Aug-17	11.60	Sigma-Aldrich Company Ltd	GOAT SERUM

CONSUMABLES	Aug-17	31.88	Sigma-Aldrich Company Ltd	TUNICAMYCIN FROM A STREPTOMYCES
CONSUMABLES	Aug-17	11.60	Sigma-Aldrich Company Ltd	GOAT SERUM
CONSUMABLES	Sep-17	-3.10	Bio-Rad Laboratories Ltd	GB VAT - GB VAT
CONSUMABLES	Sep-17	-8.82	Bio-Rad Laboratories Ltd	GB VAT - GB VAT
CONSUMABLES	Sep-17	44.08	Bio-Rad Laboratories Ltd	Mini-PROTEAN Comb 15W 1.5 mm 40
CONSUMABLES	Sep-17	-44.08	Bio-Rad Laboratories Ltd	Mini-PROTEAN Comb 15W 1.5 mm 40
CONSUMABLES	Sep-17	44.08	Bio-Rad Laboratories Ltd	Total Freight
CONSUMABLES	Sep-17	-44.08	Bio-Rad Laboratories Ltd	Total Freight
CONSUMABLES	Sep-17	-28.58	Bio-Rad Laboratories Ltd	Total Freight
CONSUMABLES	Sep-17	28.58	Bio-Rad Laboratories Ltd	Total Freight
CONSUMABLES	Sep-17	3.10	Bio-Rad Laboratories Ltd	vat q reminder sent to Juan @ Bio-Rad 17/07/17
CONSUMABLES	Sep-17	8.82	Bio-Rad Laboratories Ltd	vat q reminder sent to Juan @ Bio-Rad 17/07/17
CONSUMABLES	Oct-17	196.08	Bio-Rad Laboratories Ltd	OUT GLASS PLT W/1.5mmSP,5,M-P3
CONSUMABLES	Oct-17	112.00	VWR International Ltd	KIT ECL PRIME DET. REAGENT FOR 1
CONSUMABLES	Oct-17	252.00	Sigma-Aldrich Company Ltd	IMMOBILON-P MEMBRANE, PVDF, 0.45
CONSUMABLES	Oct-17	-75.60	Sigma-Aldrich Company Ltd	IMMOBILON-P MEMBRANE, PVDF, 0.45
CONSUMABLES	Oct-17	430.00	Life Technologies Limited	PAGERULER PLUS PRESTAINED
CONSUMABLES	Oct-17	17.64	Sigma-Aldrich Company Ltd	DULBECCOS MODIFIED EAGLES MEDIUM
CONSUMABLES	Oct-17	17.92	Sigma-Aldrich Company Ltd	RPMI-1640 MEDIUM, WITH L-GLUTAMI
CONSUMABLES	Oct-17	63.00	Sigma-Aldrich Company Ltd	cOmplete(TM), Mini, EDTA-free Pr
CONSUMABLES	Oct-17	126.00	Sigma-Aldrich Company Ltd	POLYVINYLIDENE DIFLUORIDE (IMMOB
CONSUMABLES	Nov-17	250.00	Starlab (UK) Ltd	pipette clinic
CONSUMABLES	Nov-17	-2.64	Starlab (UK) Ltd	pipette clinic
CONSUMABLES	Jan-18	44.75	Scientific Laboratory Supplies Ltd	Corning 15mL PP Centrifuge Tubes
CONSUMABLES	Jan-18	51.44	Scientific Laboratory Supplies Ltd	Falcon Conical 50ml PP Centrifug
CONSUMABLES	Jan-18	105.60	New England Biolabs	Anti-rabbit IgG HRP-linked Antib
CONSUMABLES	May-18	285.71	N/A	Transfer of expenditure PO 3718435/ Supplier Public Health England